“DO FORMER COLLEGIATE GYMNASTS MAINTAIN HIGHER BONE MINERAL DENSITY AFTER A 34-YEAR RETIREMENT FROM THE SPORT?”

by

LAUREN JEANETTE KORT

Under the Direction of Richard Lewis

ABSTRACT

The purpose of this study was to determine if the higher areal BMD (aBMD) our group previously observed in former female collegiate artistic gymnasts (GYM; n=17) compared with controls (CON; n=10), is maintained in this cohort now entering menopause (mean age 56 years). This study assessed 20-year changes in fat mass (FM), percent fat (%FAT), fat-free soft tissue, and aBMD of the total body and regional skeletal sites using dual-energy X-ray absorptiometry, as well as cross-sectional differences in trabecular and cortical volumetric bone mineral density (vBMD) and geometry at the radius and tibia using peripheral quantitative computed tomography (pQCT). Independent samples t-tests were used to determine differences between groups at baseline and at 9- and 20-year follow-ups. Repeated-measures mixed model analyses were performed to determine differences in body composition and aBMD within groups over time and to quantify the magnitude of the effects of these variables. At the 20-year follow-up, GYM vs. CON had significantly lower FM and %FAT and higher total body aBMD (all P<0.05). Over 20 years, there were group and time effects at all measured skeletal sites (P<0.05), but no group x time interactions. At the 20-year follow-up, GYM vs. CON had greater total cross-sectional area (CSA), total vBMD, bone strength index (BSI), cortical bone mineral

content (BMC), cortical CSA, and strength-strain index (SSI) at the radius, and greater BSI, total CSA, cortical BMC, cortical CSA, cortical thickness and SSI at the tibia (all $P<0.05$). These data show significant aBMD group effects at all measured sites over 20 years between GYM and CON; however, these differences were not as pronounced as they were at baseline and the 9-year follow-up. The pQCT data are in agreement with the aBMD data, and show that bone geometry differences exist between former gymnasts and controls, decades after retirement from the sport.
INDEX WORDS: GYMNASTICS, RETIRED GYMNASTS, AREAL BONE MINERAL DENSITY, BONE GEOMETRY, PAST ATHLETIC PARTICIPATION AND BONE

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DEDICATION

This thesis is dedicated to the teachers, professors, and mentors I have had in my educational career thus far. The time you have invested into me has forever touched my life.

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CHAPTER 1

INTRODUCTION

More than 40 million individuals in the United States either have already been diagnosed with osteoporosis or are suspected to be at high risk due to low bone mass. This information is troubling as approximately one in five hip fracture patients over the age of 50 years dies in the year following their fracture due to associated medical complications (National Institutes of Health Osteoporosis and Related Bone Diseases National Resource Center, NIH 2011). However, research suggests that individuals can significantly promote optimal bone health based on their lifestyle choices. Building strong bones during youth and minimizing bone loss throughout life are of principal importance to osteoporosis prevention (Heaney et al 2000).

Weight-bearing exercise, which is defined as force generating, places mechanical stress on the bone and promotes bone mineral accrual in youth and maintenance of bone mass later in life (Behringer et al 2014). Sports such as artistic gymnastics that include high-impact loading maneuvers, provide a greater osteogenic stimulus compared to other sports (Bass et al 1998; Lehtonen-Veromaa et al 2000; Modlesky et al 2008; Robinson et al 1995). Studies of collegiate artistic gymnasts versus non-gymnast controls demonstrated that gymnasts have significantly higher (8-19%) areal bone mineral density (aBMD) and favorable measures of bone microarchitecture compared to controls (Proctor et al 2001; Modlesky et al 2008). The higher aBMD observed in college gymnasts compared to other athletes (Greene et al 2012; Robinson et al 1995; Taaffe et al 1997) is most likely the accumulation of high bone mineral gains over the years of training from youth and not merely self-selection (Gruodyte-Raciene et al 2013; Laing

et al 2005). Prospective studies in prepubertal gymnasts and those entering the early stages of puberty have shown more pronounced bone mineral acquisition and benefits related to geometric and bone architecture properties in the gymnasts compared to non-gymnast controls (Laing et al 2002; Laing et al 2005; Gruodyte-Raciene et al 2013).

There are several instruments that are able to measure different properties of bone. The Dual energy X-Ray Absorptiometry (DXA) machine provides a two-dimensional image of bone and is able to quantify aBMD of the total body and regional skeletal sites (Kohrt et al 2004). Low aBMD values are considered a risk factor for skeletal fractures, which prompted the World Health Organization to use aBMD values from a DXA for diagnosis of osteoporosis. However, since DXA provides a two-dimensional image, three-dimensional bone geometry and trabecular architecture cannot be assessed. This can be problematic as larger bones may have higher aBMD than a smaller bone, purely because of difference in size rather than true bone content (Bouxsein and Seeman 2009). Computerized tomography (CT) and peripheral quantitative computed tomography (pQCT) are used to assess the bone's microarchitectural (three-dimensional) properties. Peripheral QCT images provide a measure of cortical and trabecular bone and shows muscle cross sectional area (CSA) and bone girth (Eser et al 2009). Assessment using both DXA and pQCT provides an opportunity to assess how gymnastics affects not only bone mineral content, but differences in skeletal architecture.

It has been hypothesized that the higher bone gains observed in youth are sustained into adulthood and can reduce the risk of developing osteoporosis. Cross-sectional studies of retired competitive artistic gymnasts are suggestive that bone gains are maintained into adulthood (Bass et al 1998; Ducher 2009; Kirchner et al 1996; Zanker et al 2004). Former artistic gymnasts retired from competitive training for a range of two to 20 years, were found to have significantly

higher aBMD values compared to controls at the hip (8-18%), lumbar spine (9-16%), and total body (6-9%) (Kirchner et al 1996; Zanker et al 2004). Bone geometry and microarchitecture data derived from pQCT and magnetic resonance imaging (MRI), respectively, support the DXA data. For example, former gymnasts, 18-36 years of age, who had been retired from the sport for an average of 6 years showed greater BMC, bone strength, and cross sectional area (CSA) at the radius and tibia compared to controls with differences ranging from 8% to 38% (Ducher et al 2009; Eser et al 2009). There are few long-term studies that track if bone strength and mineral gains acquired in youth persist into late adulthood. One prospective study of retired artistic gymnasts (Pollock et al 2006) showed that gymnasts in their mid-forties had significantly higher total body (9.9%), lumbar spine (11.0%), total proximal femur (7.9%), and femoral neck (11.6%) aBMD compared to non-gymnasts controls, even after a 24-year retirement from sport. The aBMD differences observed in retired gymnasts suggest that potential bone gains from athletics participation persist into adulthood. However, no long-term prospective studies have been conducted following former gymnasts into the menopausal years. Moreover, a cross-sectional study by Karlsson et al (2000) former soccer players over the age of 60 years and retired from the sport for 35 years had no residual benefits with regard to aBMD compared to controls, questioning whether the bone strength benefits of exercise training in youth and young adulthood are sustained into older adulthood. Conclusions cannot be drawn from this study, however, since the soccer players were not followed prospectively from youth into adulthood.

The purpose of the present investigation was to determine if the higher aBMD observed in former gymnasts compared to controls, previously reported by Kirchner et al (1996) and Pollock et al (2006), is still present in the same cohort of female former gymnasts 20 years later and approximately 35 years since cessation of college gymnastics training. The specific aim was

to examine changes in aBMD and related factors including body composition, physical activity, and selected nutrient intakes in the former female collegiate gymnasts and controls approximately 20 years after baseline measurements. A secondary aim was to compare the geometrical properties of bone at the tibia and radius using peripheral quantitative computed tomography (pQCT). It was hypothesized that the higher aBMD observed in former artistic gymnasts compared with controls would be maintained twenty years later as this cohort goes through menopause. Moreover, we hypothesized that bone strength will be greater in retired artistic gymnasts than controls.

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CHAPTER 2

REVIEW OF THE LITERATURE

More than 40 million individuals in the United States either have already been diagnosed with osteoporosis or are suspected to be at high risk due to low bone mass. This information is troubling as approximately one in five hip fracture patients over the age of 50 years dies in the year following their fracture due to subsequent medical complications (National Institutes of Health Osteoporosis and Related Bone Diseases National Resource Center, NIH 2011). However, some findings suggest that individuals can significantly reduce their risk for fractures and promote optimal bone strength by making healthful lifestyle choices, including participating in physical activities (Heaney et al 1995; Heaney 2000; Rizzoli et al 2010; Uusi-Rasi et al 2013). Bone-stimulating physical activities such as those associated with high impact and requiring dynamic muscular contractility may benefit the building of strong bones during youth, minimizing bone loss later in life and the progression to osteoporosis (Dolan et al 2006; Kohrt et al 2004). In this review, the following topics will be described: bone biology, assessment of bone, determinants of bone health, and exercise participation and bone health. In particular, the focus of this review will be on the effect of artistic gymnastics participation on bone in college-level artistic gymnasts, pre-menarcheal artistic gymnasts, and retired artistic gymnasts.

BONE BIOLOGY

Bone is a dynamic and metabolically active tissue that is responsible for supporting the loads applied to it. The function of the human skeleton is multifaceted. Bones function in

locomotion, act as a protector of major organs, store calcium and phosphorous, and serve as a site for blood cell formation (Anderson 2008). The skeleton is comprised of two major types of bone: cortical and trabecular. The long bones, i.e the tibia, radius, and femur, are subdivided into three regions: the epiphysis, metaphysis, and diaphysis. Trabecular bone is a soft, spongy bone that is found primarily in the ends of long bones (i.e. the epiphyses and metaphyses) and in the vertebrae. Trabecular bone has a large surface area and is very susceptible to bone turnover. Alternatively, cortical or compact bone makes up 80% of the skeleton, is found primarily in the shaft of long bones (i.e. the diaphysis), and has a turnover rate of only 3% per year (Hayden et al 1995). Osteoblasts, osteocytes, and osteoclasts are three types of bone cells that are involved in bone modeling and remodeling. Osteoblasts are bone-forming cells that produce the bone matrix, including collagen and ground substance. Osteocytes are mature osteoblasts that have been incorporated into bone matrix and provide a means of communication between cells in response to bone loading. Osteoclasts are responsible for bone resorption, or removal of old bone (Uusi-Rasi et al 2013).

Bone modeling and remodeling are processes that facilitate bone growth and maintenance throughout the lifespan. Bone modeling occurs during the growing years only and is characterized by bone formation and resorption occurring at separate anatomical sites and provides a mechanism for the skeleton to adapt geometry and size to the forces applied. Bone modeling typically occurs in females through age 16-18 years, or earlier, and in males through ages 18-20 years. Total skeletal mass peaks several years after the long bone epiphyses fuses. There is no exact age where bony accumulation plateaus due to site-specific bone differences, mainly that the hip peaks earlier than the spine, and variation in how bone mass is measured (Heaney et al 2000).

Once bone growth is completed, bone remodeling continues throughout life. Resorption of bone by osteoclasts and formation of comparable volume to osteoblasts are sequential, however, bone remodeling can be affected by age, lifestyle factors such as exercise and dietary patterns. The bone resorption phase lasts 2-3 weeks, while the bone formation stage lasts 2-3 months (Martin and Seeman 2008). Bone is renewed at a rate of 8-10% per year (Uusi-Rasi et al 2013).

ASSESSMENT OF BONE

Much of the bone is composed of inorganic materials, primarily calcium and phosphorus, but also organic materials such as collagen. It is estimated that bone is 70-90% mineral. Collagen proteins make up 90% of the remaining non-mineral components (Young 2002). Bone mass is an important determinant of bone strength. Bone size, morphometric, and material properties all contribute to overall bone strength. Research in the area of bone biology has largely focused on bone mass, as it is measurable in vivo (Heaney et al 2000), however advancements in the field have led to additional technologies capable of assessing these other important determinants of skeletal strength.

Dual energy X-Ray Absorptiometry

Dual energy X-Ray Absorptiometry (DXA) uses two X-ray beams, each with different energy levels to estimate the mineral content of the bone (Behringer et al 2014). Bone densitometry measures bone tissue absorption of photons produced by the X-ray tubes. DXA provides a two-dimensional image of bone and is able to quantify areal bone mineral density (aBMD) of the total body and regional skeletal sites (Kohrt et al 2004). Because the mass

measurement is an areal measure, when studying growing children it is important to report bone mineral content (BMC), normalized for body size, since bone volume is changing during growth and an areal measure doesn't capture "volume" in the common sense (Behringer et al 2014). Areal BMD is expressed in grams of mineral per centimeter and calculated from the BMC divided by the width of the bone at the measured site and is used to describe bone after cessation of growth throughout the remodeling process (Anderson 2008). Low aBMD values at the femoral neck and lumbar spine are considered risk factors for skeletal fractures, providing rationale for the World Health Organization to consider aBMD as the primary diagnostic criteria for osteoporosis. Using DXA as a measure of bone health can be advantageous as the machine is, for the most part, precise, provides only a low dose of radiation, is easy to use, and has a short measurement time (Bouxsein and Seeman 2009). It is important to note, however, that DXA has been known to introduce a degree of measurement error in densitometric measurements in the presence of excess soft tissue surrounding the bone (i.e. fat tissue in obese individuals). This error should be considered when DXA-derived measures are being considered in pre- and post-testing following changes in body composition because of the influence of surrounding soft tissue stores on the ability of the machine to provide an accurate densitometric measure (Yu et al 2012). However, since DXA provides a two-dimensional image, three-dimensional bone geometry and trabecular architecture cannot be assessed. This can be problematic as larger bones may have higher aBMD than a smaller bone, purely because of difference in size rather than true bone content. Although DXA has some limitations that are becoming increasingly recognized, it is considered to be the standard for assessing fracture risk (Bouxsein and Seeman 2009).

Peripheral quantitative computed tomography

Computerized tomography (CT) and peripheral quantitative computed tomography (pQCT) are used to assess the bone's architectural (three-dimensional) properties. Peripheral QCT measurements provide estimates of bone mineral content including total content, total density, trabecular content, trabecular density, cortical content, and cortical density (Zemel et al 2008). Peripheral QCT measurements also provide estimates of geometric parameters including total area, trabecular area, cortical area, cortical thickness, marrow cross-sectional area, periosteal circumference, endosteal circumference, cross sectional moment of inertia, polar moment of inertia, section modulus, strain strength index, and bone strength index (Eser et al 2009, Zemel et al 2008). In a pQCT measurement, the X-ray source rotates around the subject and uses an algorithm to construct the data into a three-dimensional image. The pQCT is calibrated with use of a bone mineral or hydroxyapatite phantom that allows for a measurement of bone density that is independent of bone size. Determination of bone geometry by use of a pQCT is clinically relevant as accelerated decreases in trabecular bone are seen with age (Bouxsein and Seeman 2009). Typically, bone is assessed at both predominantly cortical and trabecular sites. Due to the limited accessibility of the pQCT to appendicular skeletal regions, a limitation of this technology, trabecular bone is measured at the distal metaphyses whereas cortical bone is measured at the diaphysis (Zemel et al 2008). Advantages of the pQCT technology include the ability to perform a scan in a relatively short amount of time, assessment of geometric and volumetric bone density, and low radiation exposure. Although there may be an association between QCT bone density and fracture risk, practicality of using a pQCT machine instead of DXA machine to determine fracture risk remains undetermined (Bouxsein and Seeman 2009).

DETERMINANTS OF BONE HEALTH

Genetics

It is estimated that the majority (~75%) of peak bone mass is determined by genetic factors. Monozygotic and dyzygotic twins, in addition to non-twin siblings, are used for genetic comparisons and have revealed that bone mass differences between twins are linked to a region on chromosome 11 (Johnson et al 1997). Some of the key genetic factors influencing bone health are body size, hormones in the growth hormone/insulin-like growth factor (IGF) axis, vitamin D receptors, and skeletally active cytokines. In addition, genetics plays a role in nutrient utilization. For example, in individuals with similar nutrient intakes, the individual with efficient nutrient utilization will come closer to an ideal peak than the individual with inefficient nutrient utilization (Heaney et al 2000). Although there is a high level of heritability related to bone mass, other environmental factors can help to optimize bone health.

Dietary Intake

Dietary intake plays a direct role in bone tissue deposition, maintenance, and repair. Nutrients of particular importance for bone health are calcium, phosphorous, protein, vitamins C, D and K, copper, manganese, and zinc. Since the skeleton is a very large nutrient reserve for calcium and phosphorous, daily balance of absorbed intake and excretory losses of these minerals is very important (Heaney et al 1995).

Calcium is the most abundant mineral in the human body and is of vital importance to many physiological functions, including bone remodeling. Bone tissue serves as a calcium reservoir (Uusi-Rasi et al 2013). Although mild calcium deficiency does not appear to have immediate negative implications on bone, the amount of bone accumulated during growth is

partially determined by amount of calcium in the diet (Heaney et al 2000). Calcium metabolism is primarily regulated by parathyroid hormone (PTH) and 1,25-dihydroxyvitamin D (1,25-$(OH)_2D$). In older adults, net bone formation no longer balances with bone resorption and net calcium is lost from the body (Uusi-Rasi et al 2013).

The dietary reference intakes for calcium is 1,000 mg/day for women ages 19-50 years and increases to 1,200 mg/day for women ages 51-70 years (Institute of Medicine 2010). Calcium-rich foods generally include milk, yogurt and cheese. Mean calcium intake for women is approximately 800-1,000 mg/day and 60-70% of dietary calcium intake comes from milk products. Calcium supplementation may reduce the rate of bone remodeling leading to higher aBMD, but increases in bone mass occur primarily in cortical bone and the effect is most pronounced in populations with low calcium intake (Uusi-Rasi et al 2013). Variations in calcium intake early in life can make as much as a 5-10% difference in peak bone mass in adulthood. Thus, a 10% increase in peak bone density can reduce fracture risk by up to 50% in adulthood, supporting adequate calcium intake throughout the lifespan (Rizzoli et al 2010).

Phosphorus is an important component of bone mass, but is also thought to have a detrimental effect on calcium absorption. A very high dietary phosphate intake paired with a low calcium intake has been shown to cause bone loss in animal models (Heaney et al 2000). Although further research is warranted, it has been suggested that excessive phosphate consumption, usually via soft drinks, could have a negative effect on achieving peak bone mass. This could be an implication of a change in calcium balance or replacing milk with a soft drink (Heaney et al 2000).

Dietary protein provides the amino acids needed for building the bone matrix (Rizzoli et al 2010). Insufficient protein, particularly involving protein-calorie malnutrition, can cause

decreased cortical bone formation and growth retardation. Excessive protein, on the other hand, is associated with hypercalciuria in adults (Heaney et al 1995). However, since most protein-rich foods also contain phosphorus, which has a hypocalciuric effect, this effect is offset (Heaney et al 2000).

Vitamins C and K, copper, zinc, manganese, magnesium and iron may affect bone mass through their influence on bone matrix proteins. Although uncommon in the United States, deficiencies of these nutrients could lead to impairment in skeletal health (Rizzoli et al 2010).

Vitamin D

Vitamin D is a fat-soluble vitamin that plays a role in bone health by aiding calcium absorption in the intestine (Holick 2004). The updated dietary reference intakes for vitamin D are 600 IU/day for individuals ages 1-70 years. After the age of 70, the dietary reference intake increases to 800 IU/day (Institute of Medicine 2010). The main source of vitamin D is sunlight, which is converted to the active form of vitamin D by precursors in the skin. Other sources of vitamin D include fatty fish, fish oils, beef, eggs, and liver; however, vitamin D fortification of dairy foods, orange juice and cereal is becoming increasingly common (Stroud et al 2008).

Calcitriol, a form of vitamin D, regulates calcium balance in the blood. Low vitamin D concentrations can lead to hyperparathyroidism and accelerate bone loss. The optimal 25-hydroxyvitamin D concentration for optimal bone health remains under debate. What is known is that vitamin D targets intestinal, bone and kidney cells to regulate calcium balance and ensure proper bone development (Kulie et al 2009). A recent meta-analysis of vitamin D trials, without calcium, failed to show an association between supplementation and fracture prevention. However, inadequate vitamin D dose and baseline vitamin D status could have significantly skewed these results (Reid et al 2014).

Muscle

Muscle contraction places a physiological load on the bone. Harold Frost's mechanostat theory postulates that muscle force during bone growth or bone loading will affect and increase bone mass, strength, and size (Schoenau and Fricke 2008). BMC and bone strength index are a function of muscle development, and researchers have found that bone density is more or less 'constant'. Following these considerations, the functional muscle-bone unit was termed relating analyzed bone data with surrogates of muscle development and suggesting that bone mass and strength should be related to muscle function instead of age (Schoenau and Fricke 2008). With respect to muscle loss, bone atrophy precedes bone loss during unloading, but can contribute to subsequent bone loss, especially in cortical versus trabecular skeletal sites (Lloyd et al 2014, Lorentzon et al 2006). Bone strength is associated with lean (muscle) mass, particularly with BMC and area moment of inertia (Hamrick 2011). Muscle provides loads that generate bone strains and also secretes a variety of different cytokines and growth factors, myokines. Large molecules such as myostatin and vitamin D receptors can influence muscle mass and increase secretion of osteogenic myokines, thus, enhancing bone strength (Hamrick 2011). Growth and preservation of muscle mass is recommended to optimize bone health (Lloyd et al 2014).

Menstrual Disturbance

Menarche, a female's first menstrual period, is determined by genetics, hormonal factors, and many social and lifestyle variables. Primary amenorrhea is defined by absence of a menstrual cycle by age 15. It has been observed that menarche and sexual maturation occurs later in athletes, particularly gymnasts, dancers, and runners (Mountjoy et al 2014). However, Malina et al (2013) concluded that maturation of gymnasts, in particular, is similar to short late-maturing non-athletes indicating that gymnastics does not attenuate pubertal maturation.

Once menarche occurs, menstrual disturbance, especially for a prolonged period of time, can compromise bone health. Eumenorrhea, oligomenorrhea, and amenorrhea are terms used to describe menstrual status. Eumenorrhea, or normal menstrual function, is defined as cycles between 21-35 days. In adolescents, eumenorrheic cycles can range from 21-45 days. Oligomenorrhea is defined as cycle length greater than 45 days averaging out to be 3-6 menstrual cycles each year. Amenorrhea is defined as fewer than 3 menstrual cycles per year or no history of menstruation in the previous six months. Although estimates of amenorrhea prevalence vary widely, research suggests that 2-5% of college aged women experience secondary amenorrhea, while prevalence can be up to 69% in dancers and 65% in runners (Mountjoy et al 2014). Bone loss associated with menstrual disturbance may be irreversible or only partially reversible despite resumption of menstruation (Arends et al 2012). Bone loss related to amenorrhea occurs primarily in trabecular bone regions such as the spine. However, not all menstrual disturbances lead to bone loss and osteopenia. Genetics, body weight, and physical activity determine the net result of menstrual disturbance (Khan et al 2001).

Female Athlete Triad

The culture of many competitive sports, such as gymnastics, places pressure on young athletes to maintain a thin and muscular frame, which can contribute to overtraining and disordered eating behaviors (Sherman et al 1996). The combination of disordered eating behaviors and intense physical activity is generally not advantageous for bone health, as it places a female athlete at a higher risk for developing menstrual irregularities and amenorrhea (Rodriguez et al 2009). While the first two components of the female athlete triad are menstrual dysfunction and impaired bone health, the third is that of low energy availability. As defined by Loucks et al (2011), energy availability is defined by the difference between energy intake and

energy expenditure, normalized for body weight. Athletes with the 'female athlete triad' are a risk for a number of health conditions, most notably skeletal fractures. Recently, the International Olympic Committee replaced the term 'female athlete triad' to a more comprehensive term for the condition known as 'Relative Energy Deficiency in Sport' (RED-S). This term refers to the impaired physical function including, but not limited to, metabolic rate, menstrual function, bone health, immunity, protein synthesis, and cardiovascular health caused by relative energy deficiency (Mountjoy et al 2014). Research supports that adequate energy availability, as opposed to low circulating estrogen, may be the key factor in determining the body's response to physical activity. While menstrual disturbances are identified below a threshold of approximately 30 kcal/kg body weight, improving energy availability beyond this threshold should be the focus of women with exercise-induced menstrual disturbance. Reversibility of bone loss may or may not occur in athletes and depends on severity of bone loss, duration of menstrual disturbance, and treatment of menstrual disturbance (Arends et al 2012). A history of menstrual disturbance can result in compromised bone health even after restoration of menses and return to regular menstrual cycles. Restoration of BMD can be slow and may not reach the levels of age-related controls without a history of menstrual disturbance (Micklesfield et al 1995).

Menopause

Bone health outcomes in postmenopausal women are related to a combination of peak bone mass acquired earlier in life and the rate of bone loss after menopause (Mori et al 2014). Bone resorption increases with age and estrogen deficiency, both of which occur during the menopausal years. On average, menopause onset occurs at 50 years of age. At onset of menopause, estrogen production naturally and substantially declines. During menopause, there is an increase in the number of bone units that undergo remodeling and often bone resorption

outpaces bone formation (Martin and Seeman 2008). As much as 3-10% of trabecular bone can be lost during the first few years following menopause. With regard to cortical bone, approximately1% of total cortical bone can be lost in the 10 years after menopause. There appears to be a period of accelerated bone loss within the first decade of menopause, but aBMD losses levels off later in the postmenopausal years. Lifetime bone loss can be 30-40% of peak bone mass in women (Uusi-Rasi et al 2013).

Smoking

Smoking is associated with an increasing and cumulatively negative effect on bone density and leads to a greater risk of hip fracture later in life (Heaney et al 2000). Female smokers have a 17% higher risk of hip fracture at age 60 compared to their non-smoking counterparts. This effect is further increased at age 80 years, when female smokers have a 71% higher risk of hip fracture compared to females of the same age who have not smoked (Law and Hackshaw 1997).

Oral Contraceptives

A review by Tremollieres (2013) suggests that there is no clear evidence that past oral contraceptive use is associated with an increased risk of fracture. When oral contraceptive use begins within the first 3 years after menarche, however, it may interfere with bone acquisition and reduce peak bone mass. When used in adulthood, oral contraception appears to have no impact on aBMD, but could possibly prevent bone loss occurring during the perimenopausal years (Tremollieres 2013).

EXERCISE AND BONE HEALTH

Physical activity has been identified as a beneficial stimuli to bone apposition, however, not all activities are equal in terms of their osteogenic potential (Behringer et al 2014). Physical activities that expose the skeleton to high static, from impact with the ground, and dynamic, resulting from skeletal muscular contractility, loading are most advantageous in terms of bone growth (Dolan et al 2006). When an overload force is applied to bone, an adaptive response is stimulated, and continued adaptation occurs with a progressively increasing overload. Exercise stimulus to the bone causes physical deformation of the bone and subsequent adaptive response to increase bone growth (Kohrt et al 2004).

Numerous cross-sectional studies have supported the assumption that young athletes participating in high-impact sports have significantly higher BMD values when compared to their peers in non-impact sports. Deere et al (2012) performed a large cross-sectional analysis of 724 adolescents and used accelerometers to divide participants into six different impact categories, divided by impact bands ranging from 0.1-1.1*g* to >5.1*g*. Participants in high-impact bands presented with greater femoral neck BMD in boys and girls using a minimally adjusted model including age, height, and sex (0.5-1.1*g*: $\beta = -0.007$, $P = 0.81$; >5.1*g*: $\beta=0.080$, $P < 0.001$). Results suggested that high-impact physical activity, such as jumping and running, is positively related to hip BMD, while low- and moderate-impact activities provide little benefit.

Behringer et al (2014) performed a meta-analysis of 27 studies related to the effect of weight-bearing activities on BMC and aBMD during childhood and adolescence. Training program variables and characteristics were analyzed in order to better understand the relationship of exercise stimulus, individual response to exercise, and bone tissue adaptations. Subgroup analysis showed that prepubertal children have higher training-induced BMC gains when

compared to subjects who are in peripubertal or postpubertal stages, supporting the importance of pediatric exercise participation to support bone health into adolescence and adulthood (Behringer et al 2014).

Artistic Gymnastics Participation and Bone in Adults

Due to the nature of the maneuvers performed in artistic gymnastics training, gymnastics is perceived as the "gold standard" in terms of bone loading activities. Weeks and Beck (2008) developed the Bone-specific Physical Activity Recall Questionnaire (BPAQ) in order to assess the participation of research participants in activities most stimulatory to skeletal tissue. The BPAQ assigns each individual activity a specific weighting factor based predominantly upon self-measured ground reaction forces (GRFs).The weighting factors assigned to swimming (0.07), running (4.88), volleyball (31.37), aerobics (55.00), and gymnastics (100.00) vary greatly due to the perceived benefit, or lack thereof, of each individual activity in stimulating the skeleton. Due to the high level of bone loading, artistic gymnasts have sparked the interest of researchers in order to understand the influence of high impact dynamic loading on habitually loaded, as well as otherwise atypically loaded, skeletal regions.

Comparing collegiate gymnasts to non-athletes and runners, Robinson et al (1995) used DXA to examine aBMD in gymnasts (n = 21), runners (n = 20), and non-athlete matched controls (n = 19). Although gymnasts had a significantly later age at menarche and more menstrual irregularities compared to runners and controls, gymnasts had significantly higher aBMD of the total body ($P < 0.01$), lumbar spine ($P = 0.0001$) and femoral neck ($P = 0.001$) compared to runners. This study and others discussed in the following sections, suggest that aBMD is more pronounced in gymnasts than in athletes participating in other weight-bearing activities.

Greene et al (2012) examined skeletal health in a cohort of elite adolescent athletes and controls (n = 28) involved in water polo (n = 30), gymnastics (n = 25), and track-and-field (n = 34). Researchers used pQCT imaging to determine BMC, volumetric cortical and trabecular BMD, total and cortical area, and bone strength at the proximal tibia and radius. Water polo players showed 31.9% greater bone strength index at the distal radius compared to controls ($P < 0.05$). Track-and-field athletes had 33.9% greater bone strength index at the distal tibia and 14.7% greater bone strength index at the proximal tibia compared to controls ($P < 0.05$). Gymnasts, however, had the greatest musculoskeletal benefits and exhibited 60.1% and 53.4% greater bone strength index at the distal and proximal tibia, respectively, compared to controls ($P < 0.05$). Results of this study further exhibit that even at an elite level, athletes participating in other sports have limited bone gains compared to gymnasts. Researchers sought to determine the impact of exercise on bone, even in the context of menstrual dysfunction, and collegiate artistic gymnasts were identified as a population of interest.

COLLEGE-LEVEL ARTISTIC GYMNASTS

Early data recognized that weight-bearing exercise provided pronounced osteogenic benefits to athletes participating in high-impact loading sports (Nilsson and Westlin 1971). Although gymnastics is considered to have some of the highest amounts of bone loading of any sport, previous findings support that gymnasts were at high risk of developing menstrual disturbances (Sherman et al 1996). Kirchner et al (1995) reported that collegiate gymnasts consumed fewer calories and had a higher prevalence of menstrual irregularities than controls ($P < 0.05$, $P < 0.02$, respectively). The combination of reduced energy availability and menstrual disturbance is generally not advantageous for bone health (Rodriguez et al 2009). The higher

BMD observed in gymnasts, despite higher rates of menstrual disturbances, led collegiate gymnasts to become a population of interest to researchers.

Cross-Sectional Studies

In a study by Kirchner et al (1995), collegiate gymnasts without menstrual irregularities (n = 14) maintained higher aBMD than the gymnasts with menstrual irregularities (n = 7), but differences were not significant. When both groups of gymnasts were combined, the gymnasts (n = 26) had significantly higher aBMD than controls (n = 26) at the lumbar spine, proximal femur, femoral neck, Ward's triangle, and whole body (all $P < 0.0001$). These results suggest that although gymnastics participation is highly osteogenic, gymnasts with menstrual irregularities may have lower aBMD compared to gymnasts without menstrual irregularities.

Proctor et al (2002) used DXA to determine contributions of gymnastics participation on whole body, lumbar spine, and proximal femur BMD. Using a custom analysis, upper-limb BMD data was collected. Gymnasts (n = 25) had higher BMD than controls (n = 25) at all sites ($P < 0.001$). Whole body BMD was 8% higher in gymnasts compared to controls and 18-19% higher in the lumbar spine, right proximal femur, and left proximal femur. Arm BMD was 17% higher in gymnasts compared to controls. Interestingly, intragroup comparisons between the dominant and non-dominant limbs showed that controls had a significantly greater BMD in the dominant arm, but no side-to-side differences were observed in gymnasts ($P < 0.001$). Results of this study demonstrated that gymnastics training enhances BMD throughout the body, even in the non-dominant arm, while the control group showed the expected slightly decreased mineralization in the non-dominant arm.

Modlesky et al (2008) used magnetic resonance imaging (MRI) to measure microarchitectural properties of the proximal tibia in Division I female artistic gymnasts (n = 8)

and matched controls (n = 8). As evidenced by significantly higher apparent trabecular bone volume to total volume (appBV/TV) ratio of the proximal tibia, gymnasts had more optimal trabecular microarchitecture than controls. In addition, gymnasts presented with significantly higher apparent trabecular number (appTb.N; $P < 0.05$) and significantly lower apparent trabecular separation (appTb.Sp; $P < 0.05$) compared to controls, suggesting the potentially beneficial influence of gymnastics participation on trabecular microarchitecture. Results from this study helped researchers understand the effect of gymnastics participation on bone microarchitecture.

Prospective Studies

Nichols et al (1994) was one of the first researchers to follow a group of gymnasts (n = 11) throughout a 27-week competitive gymnastics season. Compared to sedentary unmatched controls (n = 11), gymnasts had significantly greater aBMD gains in the lumbar spine (1.3%), but no significant changes in the femoral neck. The short study duration of 27 weeks may not be long enough to accurately reflect the bone remodeling cycle, but these results suggest that gymnastics participation optimized bone health in the lumbar spine.

Taaffee et al (1997) examined aBMD differences between collegiate gymnasts, runners, and swimmers. The first cohort consisted of gymnasts (n = 26), runners (n = 36), and non-athletic controls (n = 14) that were followed for eight months. In this cohort, gymnasts had significantly greater increases in lumbar spine aBMD compared to runners and controls, and also exhibited significantly greater increases in femoral neck aBMD compared to controls ($P < 0.001$, $P < 0.05$, respectively). The second cohort consisted of gymnasts (n = 8), swimmers (n = 11) and non-athletic controls (n = 11) that were followed over a 1-year period. Gymnasts showed significantly greater bone gains at the lumbar spine and significantly greater changes at the

femoral neck compared to both swimmers and controls ($P < 0.01$, $P < 0.001$, respectively). Despite the short duration of these prospective studies, researchers observed greater bone mineral accrual in gymnasts compared to other athletes or sedentary controls.

PRE-MENARCHEAL ARTISTIC GYMNASTS

The culture of competitive gymnastics emphasizes a strong foundation of basic gymnastics skills that nurtures young gymnasts into elite competitors. Once early gymnastics and bone studies suggested that collegiate artistic gymnasts had higher aBMD compared to non-gymnasts, researchers sought to understand how the young starting age of gymnasts impacts bone development, if young gymnasts are predisposed to have higher BMD, and how bone accrual in young gymnasts compares to athletes in other weight-bearing sports.

Cross-Sectional Studies

Dyson et al (1997) conducted one of the earliest cross-sectional investigations of aBMD in young competitive artistic gymnasts. Gymnasts were 7-11 years of age and were required to have been training for a minimum of 15 hours per week for at least two years. Areal BMD was compared using DXA between gymnasts ($n = 16$) and controls ($n = 16$) who although were the same age as the gymnasts, were significantly taller and a higher percent body fat. Gymnasts had significantly greater femoral neck and trochanter aBMD compared to controls ($P < 0.05$). There were no significant differences in lumbar spine and total body aBMD between gymnasts and controls, most likely the result of comparing groups that were not matched for body size.

To assess aBMD of gymnasts and individually matched controls for age-, height-, and weight, Nickols-Richardson et al (2000) recruited 16 gymnasts and 16 controls ages 8-13 years. Gymnasts had been training for a mean of six years, and DXA results exhibited significantly

greater aBMD of the total proximal femur (12%; $P < 0.01$), femoral neck (14%; $P < 0.01$), trochanter (12%; $P < 0.05$), Ward's triangle (31%; $P < 0.0001$), and lumbar spine (13%; $P < 0.01$), compared to controls. These large, site-specific differences in aBMD suggest that much of the bone accrual due to gymnastics training is achieved in childhood.

Lehtonen-Veromaa et al (2000) examined if the bone mineral gain of gymnasts was different compared to athletes exposed to different loading regimens. Eleven to 17 year-old gymnasts ($n = 65$), runners ($n = 65$), and non-athletic controls ($n = 56$), were recruited for this study. Gymnasts had 20% higher aBMD of the femoral neck compared to runners and non-athletic controls ($P < 0.001$), while runners showed only 9% higher aBMD of the femoral neck compared to non-athletic controls ($P < 0.05$). Although gymnasts started training significantly earlier at a mean age of 6.6 years, and runners started training at a mean age of 8.5 years ($P < 0.001$), these finding support the hypothesis that gymnastics participation creates a more pronounced stimulus to bone compared to running.

Prospective Studies

Prospective studies support the cross-sectional evidence of the osteogenic benefits of pediatric gymnastics participation. Bass et al (1998) examined aBMD of elite gymnasts training a minimum of 15 hours per week ($n = 45$) and bone-age matched controls ($n = 35$) engaged in a minimum of two hours a week of ballet, tennis, and other bone-loading sports. Gymnasts had higher aBMD at baseline compared to controls. After 12-months, however, gymnasts accrued aBMD of the total body ($P < 0.05$), lumbar spine ($P < 0.05$), and leg ($P < 0.05$) 30-85% more rapidly than controls.

Laing et al (2002) measured the changes in BMC of 8-13 year old level 5+ gymnasts ($n = 7$) and non-gymnast controls ($n = 10$) over three years using DXA. At baseline, no initial

differences between height and weight were seen between groups. Over three years, gymnasts showed significantly greater increases in total body, trochanter, and total proximal femur aBMD and total body and lumbar spine BMD compared to controls ($P < 0.05$). Gymnasts accrued up to 30% beyond the bone gains observed in controls. Still, it was uncertain if gymnasts who have higher bone mass prior to starting any training program self-select into the sport.

In an important two-year prospective study of young 4-8 year old children who had never participated in sports, Laing et al (2005) determined that it is likely the impact loading that accounts for the higher BMC in gymnasts versus controls, not merely self-selection. Baseline measures showed that gymnasts ($n = 65$) were shorter, lighter, and had lower bone area, BMC, and aBMD compared to non-gymnasts controls ($n = 78$). Over 24-months, gymnasts had greater mean responses for total body aBMD and forearm BMC ($P < 0.04$). In addition, gymnasts had greater mean increases of lumbar spine aBMD and forearm bone area compared to controls supporting that exposure of the young skeleton to the high impact maneuvers performed by gymnasts can lead to greater bone outcomes.

Gruodyte-Raciene et al (2013) sought to determine if low-level gymnastics training influenced the estimated structural geometry development at the proximal femur. This research group followed a cohort of 165 children, categorized as gymnasts, ex-gymnasts, and non-gymnasts over the span of 4 years. DXA images of the hip were obtained annually. Gymnasts had 6% greater narrow neck cross-sectional area (CSA) than non-gymnasts (0.09 ± 0.03 cm^2, $P < 0.05$), 7% greater narrow neck section modulus (0.04 ± 0.01 cm^3, $P < 0.05$), 5% greater intertrochanter CSA (0.11 ± 0.04 cm^3, $P < 0.05$), 6% greater intertrochanter section modulus (0.07 ± 0.03 cm^3, $P < 0.05$), and 3% greater shaft CSA (0.06 ± 0.03 cm^3, $P < 0.05$). These results

show the early gymnastics exposure, even at a low-level, can provide beneficial geometric and architectural bone gains during childhood.

RETIRED ARTISTIC GYMNASTS

Both cross-sectional and prospective studies have contributed to the body of knowledge addressing whether collegiate gymnastics participation has sustained benefits into young adulthood with respect to aBMD and reduced fracture risk. What is not known is if the high aBMD associated with gymnastics training early in life is sustained into older adulthood and beyond several decades of retirement from the sport.

Cross-Sectional Studies

Kirchner et al (1996) recruited 18 former female college gymnasts, retired from gymnastics for approximately 15 years, and 15 controls matched for height, weight, and age to participate in a study at the University of Georgia. The researchers utilized DXA to compare aBMD of former gymnasts to the controls. Although height, weight, and age were matched between former gymnasts and controls, former gymnasts had significantly lower percent body fat (23.9 ± 1.0 vs. 28.8 ± 1.6%; $P < 0.02$) and greater lean mass (42.8 ± 40.9 kg; $P < 0.05$). Former gymnasts versus controls reported exercising more hours per week (16.1 ± 1.8 vs. 4.8 ± 2.0 h/wk; $P < 0.0003$) during their college years and 15 years after retirement (5.8 ± 1.2 vs. 2.9 ± 0.6 h/wk; $P < 0.05$). Even when statistically adjusting for the influence of current and past physical activity, former competitive artistic gymnasts had significantly higher lumbar spine (16%), femoral neck (18%), Ward's triangle (22%), and whole body (9%) aBMD than controls.

A few years later, Bass et al (1998) recruited 36 female gymnasts aged 18-35 years that had retired from gymnastics for a range of two to 20 years. Areal BMD was 6% to 16% higher in

former gymnasts versus controls at the total body ($P < 0.05$), femoral neck ($P < 0.01$), Ward's triangle ($P < 0.01$), trochanter ($P < 0.001$), arms ($P < 0.01$), and legs ($P < 0.01$). The significantly higher aBMD did not diminish when considering the time spent in retirement from sport.

Zanker et al (2004) examined aBMD of sedentary former artistic gymnasts ($n = 18$) and sedentary age-, height-, and weight-matched controls ($n = 18$). On average, gymnasts started training three years pre-menarche, trained until a mean age of 18 ± 2 years, and had been retired from gymnastic for between three and 12 years. The retired gymnasts had significantly higher aBMD of the total body (5.8%, $P = 0.004$), lumbar spine (9.0%, $P = 0.004$), and non-dominant femoral neck compared to controls (8.0%, $P = 0.003$). These results show that the high aBMD presumably acquired through pediatric gymnastics is still maintained into adulthood.

Using pQCT, Ducher et al (2009) reported volumetric BMD differences in 18-36 year old retired gymnasts compared ($n = 19$) to age-matched sedentary controls ($n = 24$). On average, gymnasts participated in gymnastics for 11 years and had been retired for 6 years. Retired gymnasts exhibited significantly higher bone geometric and densitometric parameters at the 4% radius ($P < 0.001$), 66% ulna ($P < 0.0001$), and 66% radius ($P < 0.001$) sites compared to controls.

Eser et al (2009) measured bone geometric and densitometric parameters on 30 gymnasts who had been retired for a mean of 6 years and 30 age-matched controls. At the radial and humeral shafts, gymnasts had higher CSA, BMC, and strength strain index (13-38%; $P \leq 0.01$) compared to controls. Total CSA and BMC were significantly greater in the distal radius, femur, and tibial shaft (8-25%) compared to controls. Former gymnasts had greater geometric

adaptations in the upper limbs compared to lower limbs, presumably due to the upper- and lower-body loading of gymnastics training.

Prospective Studies

Kudlac et al (2004) followed a group of 19 gymnasts and controls and observed changes in aBMD following a brief retirement period of four years. Baseline measurements of the gymnasts were taken at the beginning of their final competitive season, and follow-up measurements were taken after mean retirement duration of four years. At the baseline measurement, the gymnasts showed significantly higher aBMD of the femoral neck, Ward's triangle, trochanter, and total body compared to age-matched controls ($P < 0.05$). After a four-year retirement from gymnastics, former gymnasts maintained significantly higher aBMD at all skeletal sights. However, both the gymnasts and controls had significant declines in femoral neck, Ward's triangle, and greater trochanter aBMD (0.72% to 1.9% per year, $P < 0.05$). Only the gymnasts had significant declines in lumbar spine aBMD (0.87% per year, $P < 0.05$) showing that the observed benefits of gymnastics participation are not guaranteed with retirement from sport.

In 2003, Pollock et al (2006) recruited 33 participants from the cohort of gymnasts and controls that participated in the study by Kirchner et al (1996). Of the original sample, 16 former gymnasts and 13 controls agreed to participate in the follow-up study. At this time point, the former gymnasts had been retired from gymnastics for a mean of 24 years, making this the first prospective report of former gymnasts who had been retired for two-and-a-half decades. Former gymnasts continued to maintain a lower percent body fat compared to controls (25.8 ± 4.7 vs. 35.7 ± 6.6%; $P < 0.05$) and also reported greater levels of moderate physical activity than controls (1.5 ± 1.2 vs. 1.0 ± 0.6 hr/w; $P = 0.12$). Former competitive artistic gymnasts had

significantly higher total body (9.9%), lumbar spine (11.0%), total proximal femur (7.9%), femoral neck (11.6%), and forearm (13.8%) aBMD compared to non-gymnast controls (all $P<$ 0.05).

More recently, Erlandson et al (2012) examined aBMD changes of elite gymnasts (n = 25) and controls (n = 22) over fourteen years. Baseline aBMD measurements were taken on gymnasts 8-15 years old who had been involved in gymnastics training for at least two years. Premenarcheal gymnasts had significantly greater size-adjusted total body, lumbar spine, and femoral neck BMC (15%, 17%, and 12% respectively; $P < 0.05$) compared to age-matched controls. Follow-up measures were taken fourteen years later after a 6-14 year retirement from gymnastics. Gymnasts maintained similar size-adjusted total body, lumbar spine, and femoral neck BMC differences (13%, 19%, and 13% respectively; $P < 0.05$) compared with controls. Taken together, enhanced bone outcomes acquired through competitive gymnastics participation may be sustained through retirement from gymnastics.

Both cross-sectional and prospective studies have helped researchers understand how exercise, and other determinants of bone health, affects the body with respect to aBMD and reduced fracture risk. What is not known is if the high aBMD associated with gymnastics training early in life is sustained into menopause and beyond several decades of retirement from the sport. Ultimately, additional cross-sectional and long-term prospective studies of athletes and controls are warranted.

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CHAPTER 3

DO FORMER COLLEGIATE GYMNASTS MAINTAIN HIGHER BONE MINERAL DENSITY AFTER A 34-YEAR RETIREMENT FROM THE SPORT?

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ABSTRACT

The purpose of this study was to determine if the higher areal BMD (aBMD) our group previously observed in former female collegiate artistic gymnasts (GYM; *n*=17) compared with controls (CON; *n*=10), is maintained in this cohort now entering menopause (mean age 56 years). This study assessed 20-year changes in fat mass (FM), percent fat (%FAT), fat-free soft tissue, and aBMD of the total body and regional skeletal sites using dual-energy X-ray absorptiometry, as well as cross-sectional differences in trabecular and cortical volumetric bone mineral density (vBMD) and geometry at the radius and tibia using peripheral quantitative computed tomography (pQCT). Independent samples *t*-tests were used to determine differences between groups at baseline and at 9- and 20-year follow-ups. Repeated-measures mixed model analyses were performed to determine differences in body composition and aBMD within groups over time and to quantify the magnitude of the effects of these variables. At the 20-year follow-up, GYM vs. CON had significantly lower FM and %FAT and higher total body aBMD (all $P<0.05$). Over 20 years, there were group and time effects at all measured skeletal sites ($P<0.05$), but no group x time interactions. At the 20-year follow-up, GYM vs. CON had greater total cross-sectional area (CSA), total vBMD, bone strength index (BSI), cortical bone mineral content (BMC), cortical CSA, and strength-strain index (SSI) at the radius, and greater BSI, total CSA, cortical BMC, cortical CSA, cortical thickness and SSI at the tibia (all $P<0.05$). These data show significant aBMD group effects at all measured sites over 20 years between GYM and CON; however, these differences were not as pronounced as they were at baseline and the 9-year follow-up. The pQCT data are in agreement with the aBMD data, and show that bone geometry differences exist between former gymnasts and controls, decades after retirement from the sport.

Key Words: GYMNASTICS, RETIRED GYMNASTS, AREAL BONE MINERAL DENSITY, BONE GEOMETRY, PAST ATHLETIC PARTICIPATION AND BONE

INTRODUCTION

Weight-bearing exercise, which is defined as force generating, places mechanical stress on bone and promotes bone mineral accrual in youth and maintenance of bone mass later in life (Behringer et al 2014). Sports such as artistic gymnastics that include high-impact loading maneuvers, provide a greater osteogenic stimulus compared to other activities (Bass et al 1998, Lehtonen-Veromaa et al 2000, Modlesky et al 2008, Robinson et al 1995). Studies of collegiate artistic gymnasts versus non-gymnast controls demonstrated that gymnasts have significantly higher (8-19%) areal bone mineral density (aBMD) and more favorable measures of bone architecture (Modlesky et al 2008, Proctor et al 2002). The higher aBMD observed in college gymnasts compared to other athletes (Robinson et al 1995, Greene et al 2012, Taaffe et al 1997) is most likely the accumulation of high bone mineral gains over the years of training from youth and not merely self-selection (Gruodyte-Raciene et al 2013, Laing et al 2005). Prospective studies in pre-pubertal gymnasts and those entering the early stages of puberty have shown more pronounced bone mineral acquisition in the gymnasts compared to non-gymnast controls (Gruodyte-Raciene et al 2013, Laing et al 2005, Laing et al 2002).

It has been hypothesized that these higher bone gains observed in youth are sustained into adulthood and can reduce the risk of developing osteoporosis. Cross-sectional studies of retired competitive artistic gymnasts suggest that bone gains are maintained into adulthood (Bass et al 1998, Ducher et al 2009, Kirchner et al 1996, Zanker et al 2004). Former artistic gymnasts retired from competitive training for a range of two to 20 years, were found to have significantly higher aBMD values compared to controls at the hip (8-18%), lumbar spine (9-16%), and total body (6-9%) (Kirchner et al 1996, Zanker et al 2004). Bone geometry and microarchitecture data derived from peripheral quantitative computed tomography (pQCT) and magnetic resonance

imaging (MRI), respectively, support the dual-energy X-ray absorptiometry (DXA) data. For example, former gymnasts, 18-36 years of age, who had been retired from the sport for an average of 6 years showed greater BMC, bone strength, and cross sectional area (CSA) at the radius and tibia compared to controls with differences ranging from 8% to 38% (Ducher et al 2009, Eser et al 2009). There are few long-term studies that track if bone strength and mineral gains acquired in youth persist into late adulthood. One prospective study of retired artistic gymnasts (Pollock et al 2006) showed that gymnasts in their mid-forties had significantly higher total body (9.9%), lumbar spine (11.0%), total proximal femur (7.9%), and femoral neck (11.6%) aBMD compared to non-gymnasts controls, even after a 24-year retirement from sport. The aBMD differences observed in retired gymnasts suggest that potential bone gains from athletics participation persist into adulthood. However, no long-term prospective studies have been conducted following former gymnasts into the menopausal years.

The purpose of the present investigation was to determine if the higher aBMD observed in former gymnasts compared to controls, previously reported by our research team, (Kirchner et al 1996, Pollock et al 2006) is still present in the same cohort of female former gymnasts approximately 35 years since cessation of college gymnastics training. The specific aim was to examine changes in aBMD and related factors including body composition, physical activity and selected nutrient intakes in the former female collegiate gymnasts and controls approximately 20 years after baseline measurements. A secondary aim was to compare the geometrical properties of bone at the tibia and radius using pQCT. It was hypothesized that the higher aBMD observed in former artistic gymnasts compared with controls will be maintained at the 20-year follow-up as this cohort enters menopause. Moreover, we hypothesized that bone strength measured cross-

sectionally at the 20-year follow up only, will be greater in retired artistic gymnasts versus controls.

MATERIALS AND METHODS

Participants

Former competitive collegiate artistic gymnasts (n = 18) and non-gymnast controls group-matched for age-, height-, and weight (n = 15) who had no history of gymnastics participation were recruited from a local community in the southeastern United States in 1994. Participants were eligible for the parent study if they were free of physician-diagnosed bone disease or illness, were not taking medications known to affect bone development and did not have a history of smoking. Procedures were completed in this cohort at baseline (Kirchner et al 1996), approximately 9 years later (Pollock et al 2006; i.e., time point 2 in 2003-2004; 16 GYM and 13 CON), and again approximately 10 years later (i.e., time point 3 in 2013-2014 17 GYM and 10 CON; current study). Approximately 82% of participants from the baseline testing session returned for time point 3. One participant declined participation and five were not located. All returning participants were of non-Hispanic white race and signed a consent form prior to testing.

Procedures

The University of Georgia Institutional Review Board for Human Subjects approved all methods and procedures. Testing for time point 3 consisted of a single 3-hour appointment that was scheduled between September 2013 and March 2014. Testing included anthropometric measurements, DXA scans, interviewer-administered physical activity and health history questionnaires regarding medical, physical and lifestyle history. In addition, participants who

came in for testing at time point 3 also completed pQCT scans and the bone-specific physical activity questionnaire.

Anthropometric Measurements

One trained laboratory technician collected height and body weight measurements. Participants were measured for height and weight wearing light indoor clothing after removal of shoes. Anthropometric measurements were recorded three times each, and the results were averaged. Height was recorded to the nearest 0.1 cm by a wall-mounted stadiometer (Novel Products Inc, Rockton, IL). Body weight was measured to the nearest 0.1 lb by electronic scale (Seca Bella 840; Seca, Columbia, MD), and then converted to the nearest 0.1 kg. In our laboratory, one-way random effects model single measure intraclass correlation coefficients (ICCs) for height (> 0.99), weight (> 0.99) and WC (0.92) were calculated among women 18-24 years of age (N = 12) who were measured twice by the same researcher in a two-week period.

Physical Activity Assessment

Physical activity information for the past week was collected using the interviewer-administered 7-day recall questionnaire (Blair et al 1985) at time point 3. Participants reported the amount of time spent sleeping or performing moderate, hard, and very hard activities during the previous week. From this questionnaire, each participant's average daily energy expenditure (kcal/day) was estimated.

At time point 3, estimates of physical activity over the last 10 years were collected using a study-designed questionnaire developed for use in the original (baseline) study (Kirchner et al 1996). Participants were asked to list the physical activities they had participated in over the previous 10 years, and were asked about the frequency (days per week), duration (minutes each section), and intensity [1-7 (very, very easy to very, very hard)] of these physical activities. In

addition, information about gymnastics participation since cessation of competitive gymnastics training was collected.

Participants also completed a validated bone-specific physical activity questionnaire (BPAQ; Weeks and Beck 2008) at time point 3, and listed physical activities they had participated in and indicated age at participation, duration of participation, and frequency of participation. From this questionnaire, each participant received a bone-specific loading value representing their physical activity before age 15 years, after age 15 years, and over the previous year. In our laboratory, one-way random effects model single measure ICCs for 7-day PAR (0.91) and past BPAQ (0.96) were calculated among women (ages 20-22 years; N = 17) who completed the physical activity questionnaires twice in a one-week period.

Dietary Assessment

At time point 3, the Block Food Frequency Questionnaire (BFFQ; 2005 version, Berkley, CA) was used to estimate usual dietary intakes over the past year. The BFFQ has been shown to be valid and reliable for use in adults (Block et al 1990). A handout showing photographs of serving sizes was included in the questionnaire to assist participants with accurate estimations of portion sizes. Mean daily intakes of energy, protein, carbohydrate, fat, calcium, vitamin D, phosphorus, and iron were calculated.

Body Composition

Total body fat mass (FM; kg), percent body fat (%FAT), and fat-free soft tissue mass (FFST; kg) were determined using DXA (Hologic Discovery A, Hologic Inc., Waltham, MA). All scans were performed and analyzed by a single technician using Hologic Whole Body Analysis Software (version 11.2). Daily quality assurance for FM, FFST and %FAT was performed by calibration against a three-step soft tissue wedge (Hologic anthropomorphic spine

phantom, Model DPA/QDR-1; SN9374) composed of varying thickness of aluminum and Lucite, calibrated against stearic acid (100% fat) and water (8.6% fat).

Bone Outcomes

Areal BMD (g/cm^2) of the total body, lumbar spine, and non-dominant total proximal femur were acquired via DXA using guidelines from the DXA manufacturer's operator manual. All scans were performed and analyzed by a single technician using Hologic Whole Body Analysis Software (version 11.2). Daily quality assurance for DXA outcomes was performed by calibration against a three-step soft tissue wedge (Hologic anthropomorphic spine phantom, Model DPA/QDR-1; SN9374) composed of varying thickness of aluminum and lucite, calibrated against stearic acid (100% fat) and water (8.6% fat). In our laboratory women (ages 18-24 years; N = 12) were scanned by the same researcher twice in a two-week period. A one-way random effects model single measure ICCs for fat mass, FFST, %FAT and lumbar spine, total hip, femoral neck, trochanter, and radius aBMD were all ≥ 0.96.

Bone geometry was assessed using pQCT (Stratec XCT-2000; Stratec Medizintechnic GmbH, Pforzheim, Germany). A trained technician performed measurements of the non-dominant tibia and radius. Tibia scans were performed at the 4% and 38% sites of the total tibia. Radius scans were performed at the 4% and 20% of forearm length from the distal metaphysis. Image processing and calculations of the various bone indexes was determined by using the STRATEC software (version 5.50d; Stratec Medizintechnic). Each scan was obtained using a 0.4-mm voxel at a slice thickness of 2.4 mm and a scan speed of 20 mm/s. Positioning of the scans was determined in a scout view using the medial endplate as an anatomic marker and automatically set by the software at the 4%, 20%, or 38% sites. All pQCT measures were performed and analyzed by one trained operator. The pQCT operator scanned the phantom daily

to maintain quality assurance. Test-retest measurements were performed in 5 women aged 18–24 years, to determine reliability of the pQCT in our laboratory. The one-factor random effects model ICCs for all pQCT measurements were calculated to be ≥ 0.97.

Statistical Analysis

Statistical analysis was performed using the Statistical Package for the Social Sciences 21 (SPSS, Chicago, IL). Descriptive statistics were calculated to determine the range, mean, and standard deviation of all variables measured. Independent samples *t*-tests were used to determine differences between GYM and CON at baseline, during time point 2, and at time point 3 (current study). A repeated-measures mixed model analysis was performed to determine significant differences in aBMD and body composition within groups, over time and any interactions. Group differences are reported for physical activity and dietary intake for time point 3 only. Values are reported as means ± standard deviations (SD), unless otherwise noted. Statistically significant differences are reported if $P < 0.05$.

RESULTS

Participants

GYM reported to have started gymnastics training at an average age of 11.1 ± 0.9 years. Age at menarche was similar between GYM and CON ($P = 0.28$; 13.7 years ± 1.7 vs. 13.1 years ± 1.3, respectively). One GYM reported to be premenopausal and two CON reported to be going through menopause. All remaining participants reported to be postmenopausal. An additional mixed model analysis was conducted excluding the premenopausal and menopausal participants. Excluding the premenopausal and menopausal participants did not change the results (data not shown).

Participant characteristics at baseline, time point 2, and time point 3 are shown in Table 3.1. No significant differences were observed between groups in age and height at baseline, time point 2 or time point 3. At all time points, GYM had significantly lower fat mass and % body fat ($P < 0.05$) compared to CON, but no differences were observed in body weight between groups at baseline and time point 2. Both GYM and CON had significant increases in body weight ($P = 0.013$), FM ($P = 0.001$), and %FAT ($P = 0.001$), while GYM showed greater increases over time in fat-free mass compared to controls ($P = 0.045$).

Table 3.1. *Characteristics of former gymnasts (GYM) and controls (CON)*

	Baseline 1993-1994		**Time Point 2 2003-2004**		**Time Point 3 2013-2014**				
Characteristic	***GYM (n=18)***	***CON (n=15)***	***GYM (n=16)***	***CON (n=13)***	***GYM (n=17)***	***CON (n=10)***	**Group**	**Time**	**Group x Time**
Age (y)	35.8 ± 0.822	35.9 ± 0.977	44.8 ± 0.838	44.9 ± 1.089	55.8 ± 0.804	56.3 ± 1.141	-	-	-
Retirement from gymnastics (y)	15	-	24	-	34	-	-	-	-
Height (cm)	162 ± 1.40	163 ± 1.59	163 ± 1.39	163 ± 1.57	162 ± 1.42	163 ± 1.64	0.911	0.598	0.931
Weight (kg)	58.6 ± 1.53	62.6 ± 1.73	61.2 ± 1.78*	69.0 ± 2.00	64.0 ± 1.95	71.1 ± 2.26	0.013	0.000	0.121
Fat mass (kg)	13.7 ± 1.11*	18.3 ± 1.26	14.2 ± 1.06*	21.3 ± 1.19	19.9 ± 1.51*	27.1 ± 1.75	0.001	0.000	0.082
Body fat (%)	23.3 ± 1.08*	28.9 ± 1.23	23.7 ± 1.00*	30.8 ± 1.19	30.4 ± 1.27*	37.1 ± 1.50	0.000	0.000	0.614
Fat-free mass (kg)	42.3 ± 0.96	41.8 ± 1.09	43.7 ± 1.07	45.1 ± 1.21	45.4 ± 1.45	45.9 ± 1.79	0.779	0.000	0.045
aBMD (g/cm^2)									
Total body	1.165 ± 0.016*	1.091 ± 0.018	1.180 ± 0.016*	1.116 ± 0.018	1.205 ± 0.026*	1.088 ± 0.032	0.003	0.001	0.077
Lumbar spine	1.170 ± 0.026*	1.027 ± 0.029	1.185 ± 0.028*	1.047 ± 0.032	1.117 ± 0.042	0.975 ± 0.052	0.003	0.026	0.970
Proximal femur	1.030 ± 0.027	0.947 ± 0.031	1.016 ± 0.023*	0.924 ± 0.026	0.940 ± 0.026	0.867 ± .0300	0.027	0.000	0.768
Femoral neck	0.988 ± 0.030*	0.868 ± 0.034	0.945 ± 0.027*	0.841 ± 0.030	0.823 ± 0.031	0.784 ± 0.037	0.041	0.000	0.062

Values are means ± SE
* $P < 0.05$ between GYM and CON

There was a significant group effect with respect to aBMD at the total body (Figure 3.1; $P = 0.003$), lumbar spine (Figure 3.2; $P = 0.003$), proximal femur (Figure 3.3; $P = 0.027$) and femoral neck (Figure 3.4; $P = 0.041$), with GYM being higher than CON at each time point, except at time point 3 for lumbar spine, femoral neck and proximal femur. There was a

significant time effect with decreases in total body ($P = 0.001$), lumbar spine ($P = 0.026$), proximal femur ($P = 0.000$) and femoral neck ($P = 0.000$) aBMD for both GYM and CON. Although there was no statistically significant group x time interactions, there was an interaction that was approaching significance ($P = 0.062$) at the femoral neck.

Bone Geometry

Differences in bone geometry (taken at time point 3 only) of the radius and tibia are shown in Table 3.2. GYM had significantly greater measures of total cross sectional area (CSA), total volumetric bone mineral density (vBMD), and bone strength indices (BSI) at the 4% radius and significantly greater measures of total CSA, cortical BMC, cortical CSA, and strength-strain index (SSI) at the 20% radius compared to controls. At the tibia, GYM had significantly greater measures of BSI at the 4% site and significantly greater measures of total CSA, cortical BMC, cortical CSA, cortical thickness and strength strain index (SSI) at the 38% compared to controls.

Physical Activity Measures

Table 3.3 lists the questions administered using a study-designed past-physical activity questionnaire at time point 3, which pertain specifically to activity performed over the last 10 years. During the 10-year period, there were no statistically significant differences in activity between the GYM and CON with respect to frequency, duration or intensity.

Table 3.2. *Radial and tibial bone variables at the trabecular and cortical sites*

Bone Variable	GYM (n = 17)	CON (n = 9)	P value
Radius †			
Trabecular (4%) site			
Total CSA (mm^2)	317.9 ± 38.6	273.4 ± 27.6	0.000*
Total vBMD (mg/cm^3)	373.8 ± 54.7	328.9 ± 36.7	0.008*
Trabecular vBMD (mg/cm^3)	210.6 ± 36.5	190.8 ± 45.7	0.259
BSI (mg^2/mm^4)	451.1 ± 136.0	299.4 ± 73.0	0.008*
Cortical (20%) site			
Total CSA (mm^2)	130.0 ± 20.2	92.4 ± 6.9	0.000*
Cortical BMC (mg/mm)	106.4 ± 21.7	77.3 ± 10.1	0.000*
Cortical vBMD (mg/cm^3)	1186.1 ± 41.8	1166.9 ± 54.0	0.331
Cortical CSA (mm^2)	89.9 ± 20.1	66.4 ± 9.4	0.001*
SSI (mm^3)	320.9 ± 75.8	186.4 ± 23.3	0.000*
Tibia			
Trabecular (4%) site			
Total CSA (mm^2)	950.3 ± 76.7	926.0 ± 75.4	0.448
Total vBMD (mg/cm^3)	249.3 ± 31.5	235.4 ± 25.6	0.102
Trabecular vBMD (mg/cm^3)	308.7 ± 43.8	281.1 ± 28.1	0.266
BSI (mg^2/mm^4)	912.1 ± 220.1	735.2 ± 114.5	0.040*
Cortical (38%) site			
Total CSA (mm^2)	394.4 ± 41.1	349.4 ± 27.1	0.007*
Cortical BMC (mg/mm)	367.2 ± 42.8	301.4 ± 44.3	0.001*
Cortical vBMD (mg/cm^3)	1148.2 ± 43.0	1127.8 ± 37.2	0.240
Cortical CSA (mm^2)	319.9 ± 37.1	266.7 ± 33.3	0.001*
Cortical thickness (mm)	6.34 ± 0.56	5.43 ± 0.73	0.002*
SSI (mm^3)	1705.0 ± 252.7	1362.9 ± 183.9	0.002*

Values are means ± SD
† GYM n = 16 for radius measurements
* $P < 0.05$ between GYM and CON

Table 3.3. *Self-reported physical activity of former gymnasts (GYM) and controls (CON)*

Questions	GYM (n = 17)	CON (n = 10)	P Value
1. On average, how frequently have you exercised over the last 10 years (sessions/week)?	4.4 ± 3.2	4.5 ± 2.3	0.934
2. On average, how long do you exercise during each session (minutes)?	64.4 ± 17.4	55.8 ± 26.1	0.310
3. What is your intensity level of a typical exercise bout over the last 10 years [scale 1-7 (very, very easy – very, very hard, respectively)]?	4.3 ± 0.6	4.5 ± 0.8	0.463

Values are means ± SD

Current physical activity data from the 7-day physical activity recall collected at time point 3 are reported in Table 3.4. No significant differences were found between GYM and CON for hours of sleep, moderate activity, hard activity, or very hard activity. While not statistically significant, GYM had almost twice as much time spent participating in hard and very hard activities compared to CON.

Table 3.4. *Activity reported from the 7-day recall in former gymnasts (GYM) and controls (CON)*

	GYM (*n* = 17)	CON (*n* = 10)	*P* Value
Sleep (hours/wk)	7.2 ± 1.0	7.1 ± 1.6	0.959
Physical activity (hours/wk)			
Moderate	4.4 ± 4.1	5.9 ± 6.0	0.424
Hard	0.5 ± 0.7	0.1 ± 0.4	0.188
Very Hard	0.8 ± 1.0	0.3 ± 0.7	0.178

Values are means ± SD

Differences in BPAQ scores are shown in Table 3.5. GYM had significantly higher bone loading scores for < 15 years of age and for total bone loading scores than CON. There were no differences in current bone loading scores.

Table 3.5. *Bone Loading Scores in former gymnasts (GYM) and controls (CON)*

	GYM (*n* = 15)	CON (*n* = 10)	*P* Value
Bone loading score < 15 years old	155.5 ± 90.0	19.8 ± 20.4	0.000*
Bone loading score > 15 years old	135.7 ± 64.9	77.7 ± 85.9	0.067
Total bone loading score	291.2 ± 142.6	97.5 ± 96.4	0.001*
Current bone loading score	29.2 ± 33.8	21.5 ± 27.8	0.561

Values are means ± SD
* $P < 0.05$ between GYM and CON
What is the unit for BLS?

Dietary Intake

Mean dietary intakes for GYM and CON at time point 3 are reported in Table 3.6. There were no significant differences between GYM and CON for any of the nutrients reported. Both GYM and CON reported intakes of calcium that are below the dietary reference intakes of 1,000 mg/day for women ages 19-50 years and 1,200 mg/day for women ages 51-70 years (Institute of Medicine 2010). In addition, neither GYM nor CON met the Institutes of Medicine vitamin D recommendation of 600 IU/d (Institute of Medicine 2010).

Table 3.6. *Mean daily dietary intakes in former gymnasts (GYM) and controls (CON)*

Variable	GYM (n = 15)	CON (n = 10)	P Value
Kilocalories	1431.1 ± 464.8	1551.7 ± 395.3	0.508
Protein (g)	52.4 ± 17.8	62.6 ± 18.0	0.175
Carbohydrate (g)	158.1 ± 55.2	174.3 ± 51.9	0.470
Fat (g)	59.7 ± 26.6	67.5 ± 15.8	0.471
Calcium (mg)	685.3 ± 337.8	720 ± 250.5	0.783
Vitamin D (IU)	108.6 ± 84.3	102.8 ± 45.8	0.843
Phosphorus (mg)	929.0 ± 337.5	1094.8 ± 351.4	0.249
Iron (mg)	10.0 ± 4.1	12.0 ± 3.6	0.208

Values are means ± SD

DISCUSSION

The present study is the first prospective report tracking aBMD changes over 2 decades in a cohort of female former competitive gymnasts approximately 35 years following their retirement from the sport and into the menopausal years. The primary finding was that significant aBMD group effects were present at all measured sites over 20 years between GYM and CON; however, these differences were not as pronounced as they were at baseline and at the mid-point (9-year) follow-up. The pQCT data at time point 3 are in agreement with the aBMD

data, and show that bone geometry differences exist between former gymnasts and controls, decades after retirement from the sport.

At baseline, the former artistic gymnasts in this cohort had significantly higher total body, lumbar spine, and femoral neck aBMD compared to controls as described in Kirchner et al (1996). At baseline, the former artistic gymnasts had higher aBMD at the proximal femur compared to controls, but this difference was not statistically significant ($P = 0.061$). Similar results were observed in this cohort 9 years later (i.e., time point 2), as described in Pollock et al (2006). The former artistic gymnasts at time point 2 showed higher aBMD compared to controls at all measured sites including the total body, lumbar spine, proximal femur, and femoral neck. However, at time point 3, only total body aBMD was statistically different between GYM and CON, indicating that over time, the differences between groups are becoming less pronounced. We might infer based on Figure 3.4 that the former gymnasts in this study may be losing bone at the femoral neck at a faster rate than the controls; however, this was not statistically significant (group x time interaction $P = 0.062$). A longer-term follow up of this cohort is necessary to determine this.

There are several possible explanations for the significant aBMD group effect at all measurement sites in GYM vs. CON, yet a lack of statistical significance with regard to group x time interactions at the lumbar spine, proximal femur and femoral neck. It is possible that if one group begins with higher bone mass, more BMC has the potential to be lost, and possibly at a faster rate. Since no significant differences were observed between former gymnasts and controls with regard to physical activity (Table 3.4 and Table 3.5), perhaps a minimal activity level is required in adulthood to maintain aBMD. Currently there are no conclusive data on the quality

and quantity of exercise needed to preserve exercise-induced skeletal benefits in adulthood that were acquired in youth.

Although the differences in aBMD between GYM and CON are not as apparent as they were at baseline and at time point 2, cross-sectional analyses of bone geometry using pQCT showed clearly that the former artistic gymnasts had greater bone strength than controls. Ducher et al (2009) and Eser et al (2009) reported similar results between 18-36 year old retired gymnasts and controls with regard to bone geometric and densitometric parameters. The National Osteoporosis Foundation (2014) states that in postmenopausal women, such as those participating in this study, pQCT measurements of the forearm at the ultradistal radius can predict hip fracture risk. Since bone strength depends not only on the component materials, but the shape, geometry, and microarchitecture of the bone (Heaney et al 2000), the favorable bone geometry measures seen in the former artistic gymnasts suggest that this group is at a decreased risk for fracture compared to the controls.

During the menopausal years, women can lose 30-40% of their peak bone mass (Uusi-Rasi et al 2013). In this study, one participant reported to be pre-menopausal, and two controls reported to have been experiencing menopausal symptoms. Because of the rapid bone loss associated with the menopausal transition, the data were first analyzed including all participants, then analyzed again excluding the premenopausal and menopausal women. Even with these subjects excluded, there was no change in the results. However, we cannot exclude the possibility of bone changes due to hormonal changes related to menopause.

In conclusion, this study provides evidence of beneficial effects of past athletic participation on skeletal health in women going through the menopausal years. The primary finding from the present study was that there were significant aBMD group effects over 20 years

between former artistic gymnasts and controls at the total body, lumbar spine, proximal femur, and femoral neck. However, these differences were not as pronounced as they were at baseline and at the 9-year follow-up. The pQCT data are in agreement with the aBMD data, and show that bone geometry differences exist between former gymnasts and controls, decades after retirement from the sport.

ACKNOWLEDGMENTS

Funding

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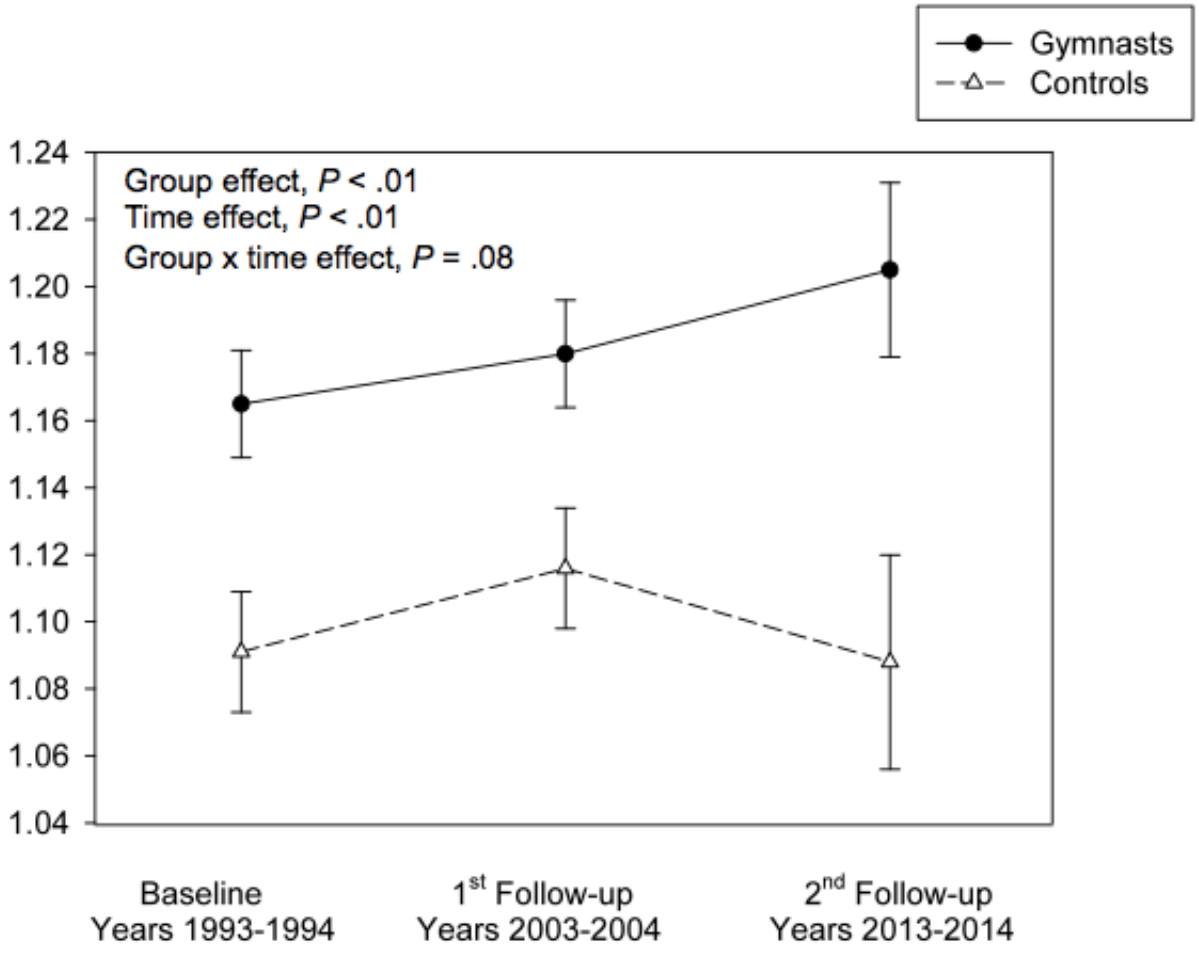

Figure 3.1. *Total body areal bone mineral density measurements between former artistic gymnasts (GYM) and controls (CON) at all time points*

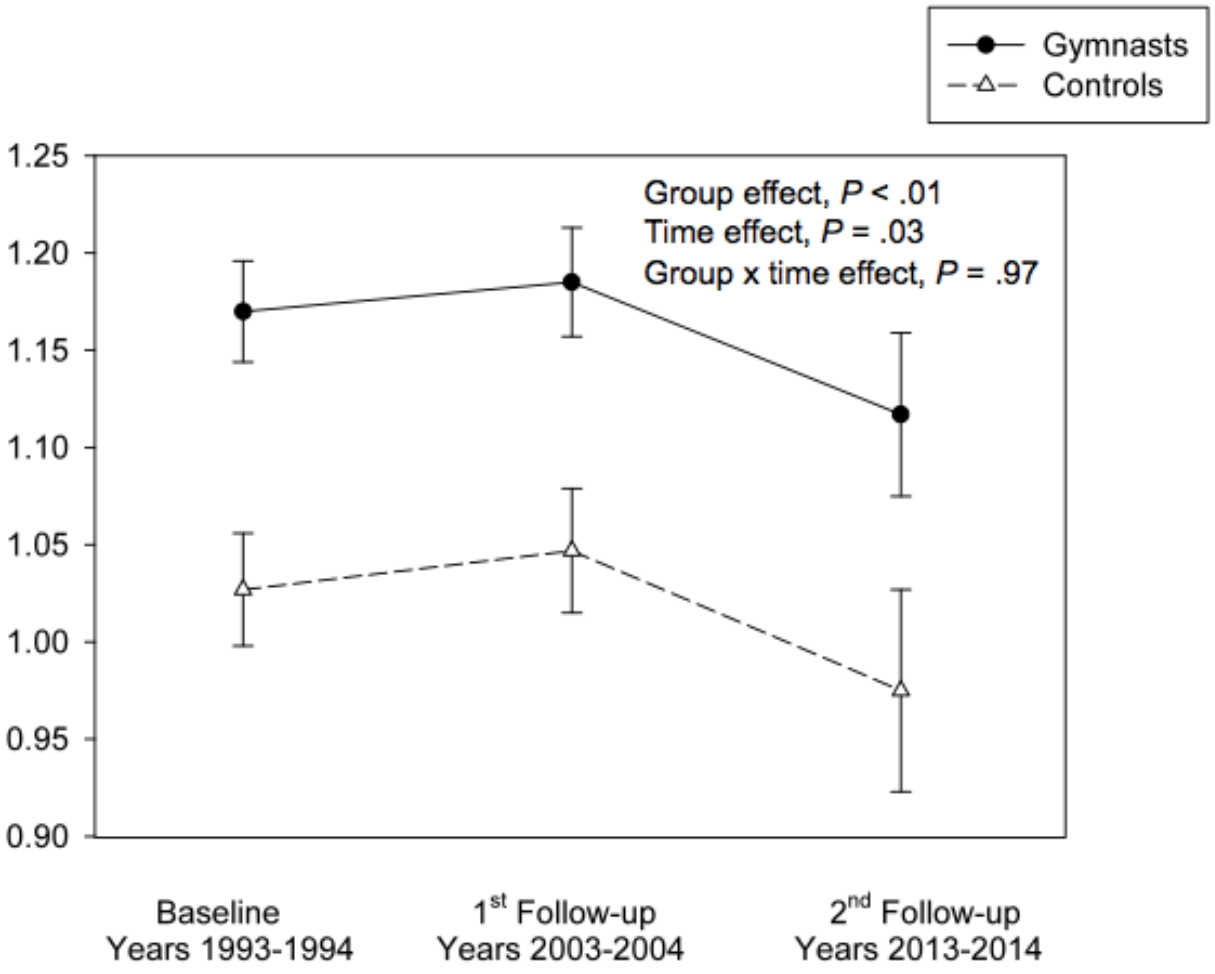

Figure 3.2. *Lumbar spine areal bone mineral density measurements between former artistic gymnasts (GYM) and controls (CON) at all time points*

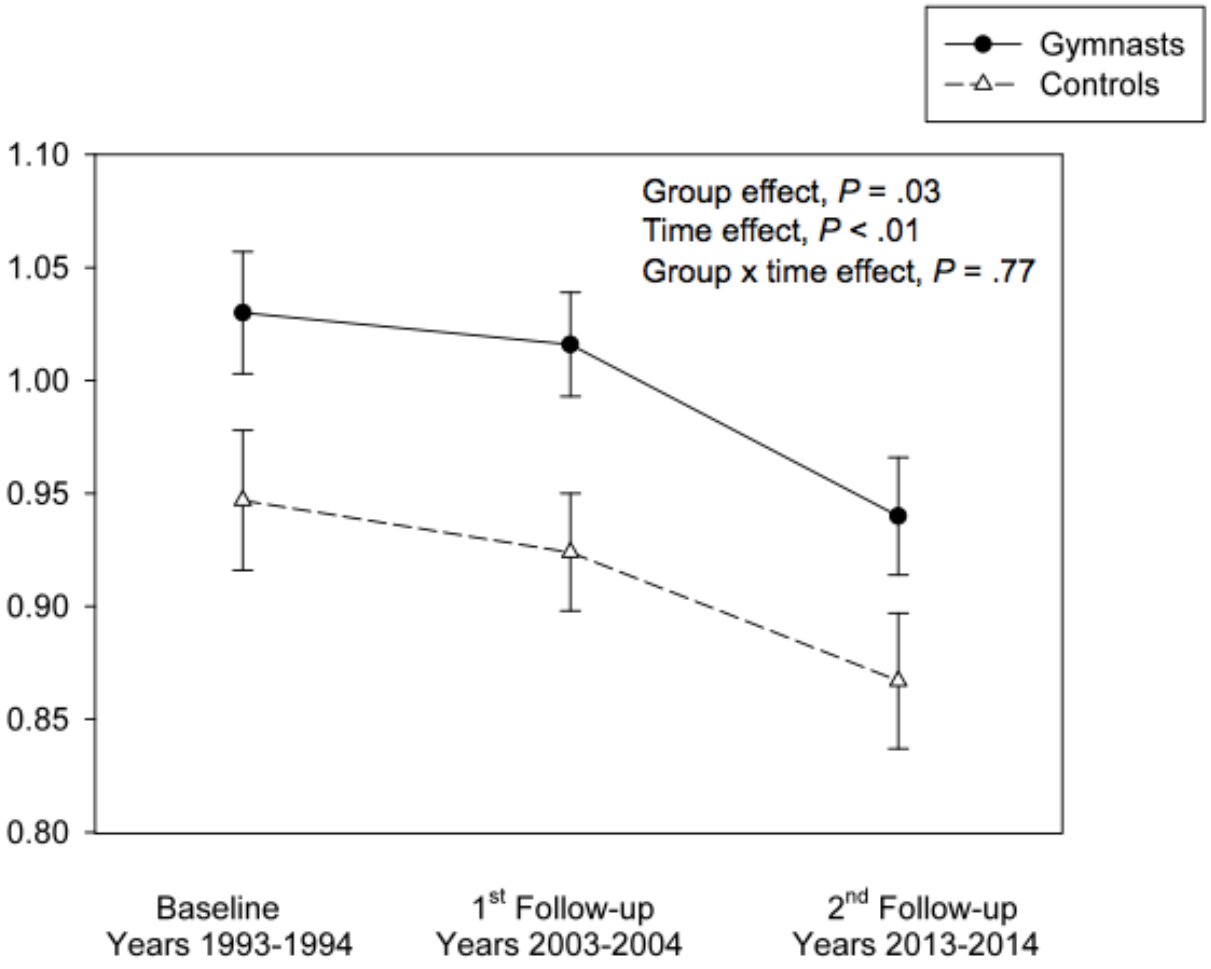

Figure 3.3. *Proximal femur areal bone mineral density measurements between former artistic gymnasts (GYM) and controls (CON) at all time points*

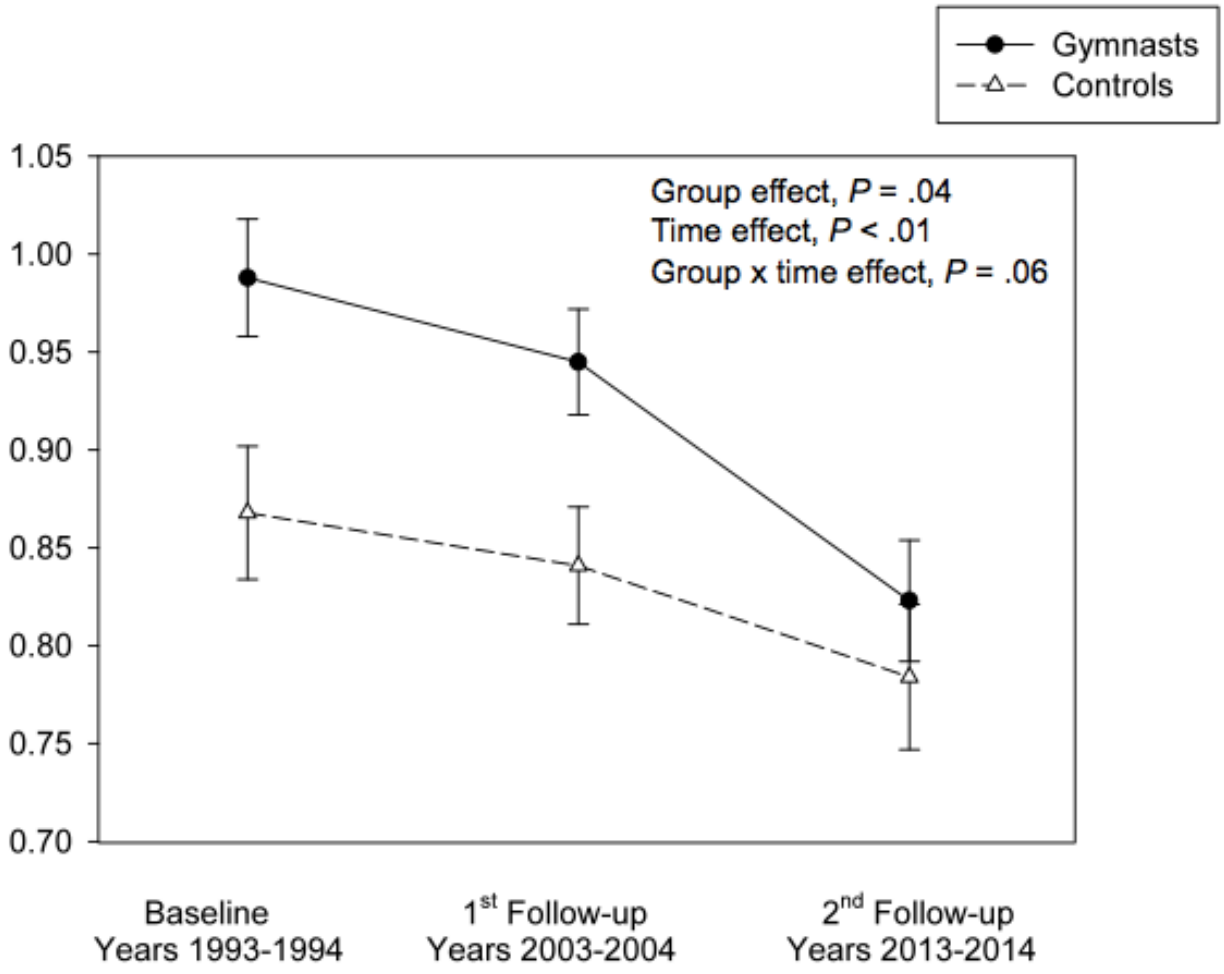

Figure 3.4. *Femoral neck areal bone mineral density measurements between former artistic gymnasts (GYM) and controls (CON) at all time points*

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CHAPTER 4

SUMMARY AND CONCLUSIONS

The present study was conducted to track areal bone mineral density (aBMD) changes over 2 decades in a cohort of female former competitive gymnasts approximately 35 years following their retirement from the sport and into the menopausal years. This study assessed 20-year changes in fat mass, percent fat, fat-free soft tissue, and aBMD of the total body and regional skeletal sites using dual-energy X-ray absorptiometry, as well as cross-sectional differences in trabecular and cortical volumetric bone mineral density and geometry at the radius and tibia using peripheral quantitative computed tomography. Information was collected regarding dietary intake, menstrual history, and current and past physical activity.

The primary finding was that significant aBMD group effects were present at all measured sites over 20 years between former artistic gymnasts and controls; however, these differences were not as pronounced as they were at baseline and at time point 2, the mid-point (9-year) follow-up. Trabecular and volumetric bone mineral density and geometry at the radius and tibia are in agreement with the aBMD data, and show that bone geometry differences exist between former gymnasts and controls, decades after retirement from the sport.

Other prospective studies in retired competitive gymnasts have observed similar results to this study; however the former gymnasts in those studies were younger and had not reached the menopausal years (Erlandson et al 2012, Kirchner et al 1996, Kudlac et al 2004, Pollock et al 2006). The present study is the first prospective report tracking aBMD changes over 2 decades in

a cohort of female former competitive gymnasts approximately 35 years following their retirement from the sport and into the menopausal years.

The results of this study are important with respect to artistic gymnastics and bone. Although baseline data for this cohort was collected approximately 15 years after cessation from collegiate gymnasts, we speculate that the higher aBMD values observed were a reflection of the training during adolescence and college, since the average start age of gymnastics training was 11 years of age. Approximately two decades after baseline measurements, we still observe those higher aBMD values although the aBMD differences between former artistic gymnasts and controls are not as apparent as they were at baseline and at time point 2. Perhaps a minimal activity level is required in adulthood to maintain aBMD. Currently there are no conclusive data on the quality and quantity of exercise needed to preserve exercise-induced skeletal benefits in adulthood that were acquired in youth.

Although the differences in aBMD between GYM and CON are not as apparent as they were at baseline and at time point 2, cross-sectional analyses of bone geometry using pQCT showed clearly that the former artistic gymnasts had greater bone strength than controls. Other cross-sectional studies in retired competitive gymnasts have observed similar results to our study with regard to bone geometric and densitometric parameters (Ducher et al 2009, Eser et al 2009).

In conclusion, this study provides evidence of beneficial effects of past athletic participation on skeletal health in women going through the menopausal years. The primary finding from the present study was that there were significant aBMD group effects over 20 years between former artistic gymnasts and controls at the total body, lumbar spine, proximal femur, and femoral neck. However, these differences were not as pronounced as they were at baseline and at the 9-year follow-up. Trabecular and volumetric bone mineral density and geometry data

are in agreement with the aBMD data, and show that bone geometry differences exist between former gymnasts and controls, decades after retirement from the sport.

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APPENDICES

APPENDIX A

Consent Form

UNIVERSITY OF GEORGIA
CONSENT FORM
The Long Term Effect of Gymnastics Participation on Bone Health

Researcher's Statement
I am asking you to take part in a research study. Before you decide to participate in this study, it is important that you understand why the research is being done and what it will involve. This form is designed to give you the information about the study so you can decide whether to be in the study or not. Please take the time to read the following information carefully. Please ask the researcher if there is anything that is not clear or if you need more information. When all your questions have been answered, you can decide if you want to be in the study or not. This process is called "informed consent." A copy of this form will be given to you.

Principal Investigator: *Dr. Richard Lewis*
Department of Foods and Nutrition
706-542-4901/ rlewis@fcs.uga.edu

Purpose of the Study
The purpose of the study is to determine how participation in competitive gymnastics during college may affect bone mineral density later in adulthood. You are being asked to participate because of your prior involvement in this research.

Study Procedures
If you agree to participate, you will be asked to:

- *Following an overnight fast, have blood drawn by a qualified and experienced phlebotomist who will follow standard sterile techniques, monitor you after the needle is withdrawn, and apply pressure to the blood-draw site. Approximately six teaspoons (30mLs) of blood will be drawn. After the blood draw, you will be given a snack. You do not have to give a blood sample. If you choose not to give blood, you can still be in this study.*
- *Complete a health history questionnaire, past physical activity questionnaire, bone-loading questionnaire, food frequency questionnaire and an interviewer- administered 7-day physical activity recall. The approximate total time to complete all questionnaires will be approximately 45 minutes.*
- *Your height and body weight will be measured.*
- *Complete the bone density scans of the total body, lumbar spine, forearm and hip on the Discovery A bone scanning (DXA) machine, and scans of the non-dominant leg and forearm with a 3-dimensional bone scanning (peripheral QCT) machine. The total time for completion of all scans will be approximately 60 minutes.*
- *Someone from the study may call you if any of your information needs clarification.*
- *Your individually-identifiable information will be kept for ten years. This will allow the researchers to contact me should there be another similar follow-up study in the future, at which time you can decide if you wish to participate again.*

Approved by University of Georgia
Institutional Review Board
Protocol # MODCR00000095
Approved on: 9/19/2013
For use through: 9/18/2014

Risks and discomforts

- *The risks of participating in this study are minimal:*
  - *Psychological risks: The discomforts or stresses that you may face during this research include psychological discomfort from the disclosure of individually-identifiable information concerning diet, physical activity and history of menstruation status. However, you may skip any question that may be distressing. If undue discomfort or stress occurs, you have the right to discontinue the study at any time.*
  - *Physical risks:*
  - *Blood Draw: You may experience some discomfort or stress when your blood is drawn. The risks of drawing blood from your arm include the unlikely possibilities of a small bruise or localized infection, bleeding, and fainting. These risks will be reduced in the following ways: your blood will be drawn only by a qualified and experienced person who will follow standard sterile techniques, who will observe you after the needle is withdrawn, and who will apply pressure to the blood draw-site. If a blood sample cannot be obtained after two attempts, no further attempts will be made.*
  - *Bone Scans: You will be exposed to a small amount of radiation during the bone (DXA and pQCT) scans. The DXA and pQCT scans performed will total 140 µSv of radiation. For comparison, natural and man-made background exposure is approximately 122µSv per week (source, US EPA). The total amount of possible radiation is far less than the 500 to 800 µSv of radiation received from an adult chest X-ray. Alternatively, a round-trip airline flight from Athens, GA to Athens, Greece would be approximately 140µSv of exposure, equal to the total amount of radiation exposure. Considering these comparisons, it is reasonable to assess the risk of harm from the amount of radiation exposure for subjects versus non-subjects as minimal. In the event that information from any scan is lost or unusable, no additional scans will be performed. A copy of your bone scans will be provided to you, but the researchers are not medical doctors. The DXA results will be explained to you, and may be clinically relevant, but for diagnosis and health questions, you should consult a qualified physician.*
  - *Pregnancy Risks: Being a part of this study while pregnant may expose the unborn child to a yet undiscovered risk. Therefore, pregnant women or those who suspect they could be pregnant will be excluded from the study. By signing this form, females of childbearing potential are certifying to the best of their knowledge that they are not pregnant and agree to utilize adequate birth control methods during their participation in this study. If you express any doubts regarding pregnancy status, a pregnancy test will be provided to you, which you may complete in a private location prior to undergoing DXA or pQCT scanning. If the pregnancy test is positive, you may maintain confidentiality by electing not to disclose any information to the research group, but must voluntarily decline to take the DXA or pQCT tests. If you elect to notify the research group of the pregnancy, you will receive information about and referral to an OBGYN. Refusal to take the pregnancy test will also be documented below.*

I certify that I am not pregnant, or trying to become pregnant.
(Check one): ***YES____*** ***NO____***

I was given the opportunity to complete a simple urine test for pregnancy.
(Check one): ***YES____*** ***NO____***

I understand the risks described above, and refuse to take the pregnancy test.
(Check one): ***YES____*** ***NO____***

Benefits

- *The benefits you can expect from participation are the assessment of bone health (bone mineral density and bone mineral content), body composition (percentage of body fat and non-fat tissue), diet and physical activity patterns.*
- *The researcher also hopes to learn more about the impact of competitive gymnastic exercise throughout childhood, adolescence and early adulthood on bone health after retirement from sport.*

Incentives for participation

If you are an out-of-town participant, you will be reimbursed for travel expenses for up to $400. In addition, the value of the DXA bone and body composition scans is estimated to be about $800.

Privacy/Confidentiality

Although some individually-identifiable information will be collected from you for contact and payment purposes, all data collected as part of the study procedures will be coded using an identifying subject number. The key to the code will be kept in the researcher's offices under lock and key, or in a password protected computer file. Only the researcher and members of this research team will have access to identifiable data. The project's research records may be reviewed departments at the University of Georgia responsible for regulatory and research oversight. The key to the code matching your name with your ID number will be destroyed following a ten-year retention period.

Researchers will not release identifiable results of the study to anyone other than individuals working on the project without your written consent unless required by law.

Taking part is voluntary

Your participation is voluntary. You can refuse to participate or stop taking part at anytime without giving any reason, and without penalty or loss of benefits to which you are otherwise entitled.

If you decide to withdraw from the study, the information that can be identified as yours will be kept as part of the study and may continue to be analyzed, unless you make a written request to remove, return, or destroy the information.

If you are injured by this research

The researchers will exercise all reasonable care to protect you from harm as a result of your participation. In the event of an injury as an immediate and direct result of participation, the researchers' sole responsibility is to arrange for transportation to an appropriate facility if additional care is needed. If you think that you have suffered a research-related injury, you should seek immediate medical attention and then contact *Dr. Richard Lewis* right away at *706-542-4901.* In the event that you suffer a research-related injury, your medical expenses will be your responsibility or that of your third-party payer, although you are not precluded from

seeking to collect compensation for injury related to malpractice, fault, or blame on the part of those involved in the research.

If you have questions

The main researcher conducting this study is *Dr. Richard Lewis*, a *Professor and Principal Investigator* at the University of Georgia. Please ask any questions you have now. If you have questions later, you may contact *Dr. Richard Lewis* at *rlewis@fcs.uga.edu* or at *706-542-4901*. If you have any questions or concerns regarding your rights as a research participant in this study, you may contact the Institutional Review Board (IRB) Chairperson at 706.542.3199 or irb@uga.edu.

Research Subject's Consent to Participate in Research:

To voluntarily agree to take part in this study, you must sign on the line below. Your signature below indicates that you have read or had read to you this entire consent form, and have had all of your questions answered.

_____	_____	_____
Name of Researcher	Signature	Date
_____	_____	_____
Name of Participant	Signature	Date

Please sign both copies, keep one and return one to the researcher.

Approved by University of Georgia
Institutional Review Board
Protocol # MODCR00000095
Approved on: 9/19/2013
For use through: 9/18/2014

APPENDIX B

Anthropometric recording sheet

The Long Term Effect of Gymnastics Participation on Bone Health

Participant Information Sheet
Anthropometrics/DXA/pQCT

Subject ID: _____ Visit Date:_____

DOB: Month _____ Day _____ Year _____

Weight (lb): _____ _____ _____ _____

Measure 1 Measure 2 Measure 3 Average

Height (cm): _____ _____ _____ _____

Measure 1 Measure 2 Measure 3 Average

Forearm Length (cm): _____ _____ _____

Measure 1 Measure 2 Average of 1 and 2

BMI (g/cm²): _____

Non-Dominant Arm: R L circle one
Non-Dominant Leg: R L circle one

DXA operator use	PQCT operator use
☐ Total Body ☐ Lumbar Spine ☐ Hip ☐ Forearm Scan date: _____ Completed by: _____ initials of operator	☐ Arm Length _____ 66% length _____ ☐ Leg Length _____ 66% length _____ Scan date: _____ Completed by: _____ initials of operator

APPENDIX C

Health history questionnaire

Name:_____ Age:_____ Date of Birth:_____
Address:_____ Home Phone #: _____ Work Phone #:_____
_____ Occupation:_____ Cell Phone #:_____
Race/Ethnic Background:_____

Child Bearing History

1. How any children have you given birth to?
   _____
2. What are their ages?
   _____
3. Did you breast-feed any of you children? _____ yes _____ no. If yes, how long?
   _____
4. Did you have any C-sections? _____ yes _____ no

Menopausal History

1. Have you gone through menopause (12 months without a period)? _____ yes _____ no. If yes, how old were you when it occurred?
   _____
2. Are you presently going through menopause? _____ yes _____ no. If yes, how long ago did you start going through it?
   _____
3. Are you using any medications relating to your menopause? _____ yes _____ no. If yes, which medications?
   _____
4. When did you start using these medications?
   _____

Surgery/Medication History

1. Please list major medical procedures, surgeries and/or injuries in your lifetime and related medications. Give the time of the procedure or injury and/or the frequency and duration of medication.
   _____
   _____
   _____

_____
_____

2. Have you ever gone through an extended period of time where you were bedridden or immobilized? _____ yes _____ no. If yes, how old were you and how long did this immobilization last? Briefly explain the circumstances.
_____
_____

Other History

1. Do you smoke cigarettes now? _____ yes _____ no. If yes, on average, about how many cigarettes a day do you smoke now? _____1-5, _____6-14, _____15-24, _____25-35, _____35 or more
2. If you used to smoke but do not smoke now, how long did you smoke? _____ years. On the average, about how many cigarettes a day did you smoke? ____1-5, ____6-14, ____15-24, ____25-35, ____35 or more
3. How old were you when you began using birth control pills (if ever used)? _____ How long have you been using them?
_____
4. What periods of time did you stop using birth control polls? (Please give dates, if applicable)
_____
_____
_____
5. How would you rate your present health? _____ Poor _____ Good _____ Fair _____ Excellent
6. Any history of bone diseases? _____ yes _____ no. If yes, explain?
7. Are your menstrual cycles regular? _____ yes _____ no. If not, how long have they been irregular? When was your most recent period?
8. Any significant weight changes in the last ten years?
9. Are you on any nutritional supplements?
10. Are you currently dieting or on any special type of weight loss program (Weight Watchers, Atkins, etc…)
11. Has any member of your family been diagnosed with osteoporosis?
12. Do you have any health problems that limit your physical activity?
13. How many hours, on average, do you spend watching TV or on the computer?

APPENDIX D

Seven-day physical activity questionnaire

Subject ID #: _____
Interviewer: _____
Date of Interview: _____

7-day Physical Activity Recall Questionnaire
Physical Activity List

Moderate Activities

Occupational Tasks:

1. Delivering mail or patrolling on foot
2. House painting
3. Truck driving (making deliveries – lifting and carrying light objects)

Household activities:

1. Raking the lawn
2. Sweeping and mopping
3. Mowing the lawn with a power mower
4. Cleaning windows

Sports Activities (Actual playing time):

1. Volleyball
2. Ping Pong
3. Brisk walking for pleasure or to work (3 mph or 20 min/mile)
4. Golf-walking and pulling or carrying clubs
5. Calisthenic exercises

Hard Activities

Occupational Tasks:

1. Heavy carpentry
2. Construction work – doing physical labor

Household Tasks:

1. Scrubbing floors

Sports Activities (Actual playing time):

1. Double tennis
2. Disco, square, or folk dancing

Very Hard Activities

Occupational Tasks:

1. Very hard physical labor – digging or chopping with heavy tools

2. Carrying heavy loads, such as bricks or lumber

Sports Activities (Actual playing time):

1. Jogging or swimming
2. Singles tennis
3. Racquetball
4. Soccer
5. Aerobics
6. Stair climbing
7. Weight training
8. Gymnastics

1. On the average, how many hours did you sleep each night during the last 5weekday nights (Sunday-Thursday)? Record to the nearest quarter-hour.
   Hours: _____ Minutes: _____

2. On the average, how many hours did you sleep each night last Friday and Saturday nights?
   Hours: _____ Minutes: _____

3. First, let's consider moderate activities. What activities did you do and how many total hours did you spend during the last 5 weekdays doing these moderate activities or others like them? Please tell me to the nearest half-hour.
   Hours: _____ Minutes: _____

4. Last Saturday and Sunday, how many hours did you spend on moderate activities and what did you do? (Can you think of any other sport, job, or household activities that would fit in this category?
   Hours: _____ Minutes: _____

5. Now let's look at hard activities. What activities did you do and how many total hours did you spend during the last 5 weekdays doing these hard activities or others like them? Please tell me to the nearest half-hour.
   Hours: _____ Minutes: _____

6. Last Saturday and Sunday, how many hours did you spend on hard activities and what did you do? (Can you think of any other sport, job, or household activities that would fit in this category?)
   Hours: _____ Minutes: _____

7. Now let's look at very hard activities. What activities did you do and how many total hours did you spend during the last 5 weekdays doing these very hard activities or others like them? Please tell me to the nearest half-hour.
   Hours: _____ Minutes: _____

8. Last Saturday and Sunday, how many hours did you spend on very hard activities and what did you do? (Can you think of any other sport, job, or household activities that would fit in this category?)
   Hours: _____ Minutes: _____

9. Compared with your physical activity over the past 3 months, was last week's physical activity more, less, or about the same? (Circle one)
   More Less About the same

APPENDIX E

Past physical activity questionnaire

I.D._____

PHYSICAL ACTIVITY QUESTIONNAIRE

1. On the average, over the last 10 years, how frequently have you exercised (including going on walks, riding a bicycle, dancing, etc.)? Report the total number of times that you have exercised in a typical week.

   _____

2. On the average, how long do you exercise each time? _____
3. Circle the number that best represents the intensity of your typical exercise bout:
   1. Very, very easy
   2. Very easy
   3. Easy
   4. Average
   5. Hard
   6. Very hard
   7. Very, very hard

4. What specific physical activities have made up your exercise routine? How long have you spent doing each activity?

   _____
   _____
   _____

5. During your college years, how frequently did you exercise (including going on walks, riding a bicycle, dancing, etc…) Report the total number of times that you exercised in a typical week.

   _____
   _____
   _____

6. On the average, how long did you exercise each time? _____
7. Circle the number that best represents the intensity of your typical exercise bout:

For former gymnasts:

8. Since you have stopped competing in gymnastics, have you done any gymnastics activities (uneven bars, vault, etc)? _____ yes _____ no

If yes, what activities, when, and how long?

_____
_____
_____

APPENDIX F

Bone loading history questionnaire

Bone-Specific Physical Activity Questionnaire (BPAQ)

SUBJECT ID:	DATE:

1. Please list <u>any</u> sports or other physical activities you have participated in regularly. Please <u>tick</u> the boxes to indicate how old you were for each sport/activity and how many years you participated for.

Activities \ Age:	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30

Activities \ Age:	31	32	33	34	35	36	37	38	39	40	41	42	43	44	45	46	47	48	49	50	51	52	53	54	55	56	57	58	59	60

Activities \ Age:	61	62	63	64	65	66	67	68	69	70	71	72	73	74	75	76	77	78	79	80	81	82	83	84	85	86	87	88	89	90

BONE-SPECIFIC PHYSICAL ACTIVITY QUESTIONNAIRE
Developed by B.K. Weeks and B.R. Beck
Griffith University, QLD, Australia

Bone-Specific Physical Activity Questionnaire (BPAQ)

SUBJECT ID:	DATE:

2. Please list the sports or other physical activities (be as specific as possible) you participated in regularly during the <u>last 12 months</u> and indicate the average frequency (sessions per week)?

Activity: _____ Frequency (per week): _____

Activity: _____ Frequency (per week): _____

Activity: _____ Frequency (per week): _____

Activity: _____ Frequency (per week): _____

Activity: _____ Frequency (per week): _____

Activity: _____ Frequency (per week): _____

Activity: _____ Frequency (per week): _____

Activity: _____ Frequency (per week): _____

BONE-SPECIFIC PHYSICAL ACTIVITY QUESTIONNAIRE
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