PROTEIN INTAKE AND SITE SPECIFIC BONE MINERAL DENSITY
IN CAUCASIAN MALE RESISTANCE TRAINERS

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Thesis

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CHAPTER I
THE PROBLEM

Introduction

Resistance trainers are typically a group of people that use diet as a tool for athletic development. Many resistance trainers partake in consuming a high protein diet to aid in muscle growth. A stereotype of the athletes participating in strength sports is that they will do anything to succeed, without concern for the health of their body. Many do not know the safety or efficacy of excess protein on their body, such as their bone mineral density (BMD).

Statement of the Problem

High protein diets have been met with controversy over the safety of the diet for the overall health of bones. Various researchers believe that diets high in protein can cause lower BMD, [1] while others state that excess protein in the diet does not harm BMD [2, 3]. The primary purpose of this study was to research the effects of high protein diets on site-specific BMD in a lesser-studied segment of the population, Caucasian male resistance trainers. The authors hope to add their findings to current literature related to the safety of consuming excess protein.
Motivation

Previous research has been focused on the potentially harmful aspects of excess protein on bone health, largely based on the calciuria induced by particular amino acids. However, recent studies are showing no deleterious effects on actual whole-body bone mineral density. Examining specific skeletal sites that are commonly used to assess BMD may reduce some of the limitations of whole-body DEXA scans (e.g. large individuals) and provide more information.

Importance

Research has, to present, never been performed on this population with a multi-year duration of protein consumption. This study was the first to document extreme protein intakes over a long duration in relation to bone mineral density, especially in a population that is known to consume ample protein. This research presents data on a topic that is relative to the population studied and provides information to add to scientific literature.

Goals and Hypothesis

We aim to present evidence that high protein diets are safe for maintenance of site-specific bone mineral density, specifically in Caucasian male resistance trainers. Our hypothesis is that protein-seeking Caucasian male resistance trainers will not differ from non-protein-seeking peers in site-specific BMD, i.e. femoral neck and lumbar spine.
Definitions

**Bone Mineral Density (BMD)** - a measurement of the density of the bones in grams per square centimeter (g/cm²) that represents inorganic content within the bone, such as calcium, phosphorus, and magnesium.

**Dietary Protein** – Whole food and dietary supplements including casein, whey, egg, meat and soy.

**Dual x-ray absorptiometry (DEXA)** – an assessment tool used to analyze bone mineral density. DEXA is the most common and preferred measurement tool for assessing BMD because of its precision and low subject exposure to radiation. DEXA uses laser x-ray scanners and specific computer software to assess the risk of fractures, measured in standard deviations, from comparison of bone mass of the patient to that of a healthy person of similar age and sex [4].

**Femoral Neck** – Area on the superior aspect of the femur, near the pelvis, located between the greater trochanter and femoral head [5].

**Lumbar Spine** – Vertebrae located at the base on the spine below the thoracic and cervical vertebra. It is also referred to as the lower back.

**Newton (N)** – A force, which, if applied for one second, will cause a one-kilogram object starting from rest, to reach a speed of one meter per second [6].

**Osteopenia** – Literal translation from Greek means, “bone poverty”. It is considered a midway condition in which bones are weakened, and there is an increased fracture risk [7]. Osteopenia occurs when bone density is between 2.5 and 1.0 standard deviations below normal for age and gender.
**Osteoporosis** – Literally translated means “porous bones”. Osteoporosis occurs when bone density is more than 2.5 standard deviations below what is normal for age and gender [7].

**Rate of Perceived Exertion (RPE)** – Intensity of exercise as measured by the individual performing the activity. Based on the Borg scale, measurements range from six to twenty, with six being no exertion at all and twenty being maximum exertion.

**Resistance Athletes** – Athletic persons who identify themselves as focusing on muscle mass and strength and, on average, lift three or more days per week. (i.e. power lifters, body builders, Olympic lifters, strongman athletes).

**Assumptions**

The following assumptions are made in regard to this study:

1. Participants will record their diets accurately and honestly
2. Participants will record their exercise program accurately and honestly

**Limitations**

A limitation of this study is that it was semi-observational in the fact that the researchers are only collecting information from the participants and not intervening in their everyday diet. The only thing that was controlled was activity level at specific times during the study (i.e. pre-testing). Another limitation was that diet logs may not be completed as accurately as was needed to optimally analyze them to obtain the most precise data. This study was also a retrospective study. Our analysis was done using data from a previous related study.
CHAPTER II
REVIEW OF THE LITERATURE

Introduction

The question concerning liberal protein consumption (a high protein diet) and safety, specifically bone health, has been a controversial issue in the fields of dietetics, athletics, and sports medicine. This study will attempt to ascertain whether high protein diets are safe and healthy for active individuals, namely resistance trainers, who wish to partake in consuming ample protein. A negative result (i.e. no deleterious impact on BMD) would refute what many educational sources [2] and some scientific sources [1] of literature state as fact, that a diet too high in protein is unhealthy and possibly unsafe, particularly for bone health. The present study would enhance recent literature that has shown no relationship between large protein intakes and harmful results.

Population

Resistance trainers, stereotypically, fit into a group of people who are more likely to attempt to consume extra protein [3, 8]. Protein is seen as an essential tool for success to help increase muscle mass and gain strength. The thought is that extra protein will increase protein synthesis in muscle tissue and lead to an increase in muscle mass. As evidenced by a worldwide protein supplement industry, numerous resistance trainers add
extra protein to their typical diet to support their resistance workouts in the gym and improve muscle mass.

Resistance trainers are a segment of the population that has not been studied as thoroughly as other sections of the population. These athletes are an ideal group to study regarding protein intake and bone mineral density (BMD) because the body of a resistance trainer displays an extreme example of the capability of the human body. By looking at the maximal capabilities of the human body, more can be learned and applied to other segments of the population.

_Bone Mineral Density and Resistance Athletes_

It is known that BMD is a measurement of the density, and by extension strength, of the bones, that is representative of mineral content. It can be influenced by genetic or environmental factors [9,10]. Measurement for BMD is typically measured in g/cm² and can be measured as either whole body or site-specific BMD. Site-specific BMD, such as at the lumbar spine and femoral neck, can differ from whole-body measures, but more often, a correlation is seen between the two measures.

Athletes that partake in resistance training have a higher BMD [10,11]. Resistance athletes show an increase in both bone resorption and formation markers indicating that there is a high level of bone remodeling occurring. Sedentary individuals exhibit an increase in bone resorption markers without an increase in bone formation markers [10]. This indicates that bone loss supersedes bone development.

Karlsson et al. (1993), showed that weight lifters have a higher BMD in, not only total body, but also in the lumbar spine and the greater trochanter region [11]. It has
previously been shown that a greater body mass has been associated with a higher total-body and femoral neck BMD [9]. A previous participation in sports has also been reported to enhance BMD in the lumbar spine region [9]. The opposite phenomenon was exhibited to be true with lesser participation in sports during the subject’s life, leading to lower lumbar spine BMD [9].

In a study of powerlifters competing in national competitions, it was shown that the average compressive loads on the L4/L5 vertebrae were upwards of 17,192 N during dead lifts [12]. From these data, it can be said that the lumbar spine experiences approximately 3,900 pounds of force when performing this exercise. The bones of a resistance trainer are called upon to bear heavy loads and it is known that weight-bearing exercise increases BMD [9, 10, 13, 14].

Protein

Proteins are made up of amino acids linked together by peptide bonds. There are nine amino acids, termed essential amino acids, which the body is unable to synthesize and must therefore be taken in through in the diet. Eleven amino acids are called nonessential because the body is able to manufacture them and there is no need for them to be consumed through the diet [7].

There are two specific types of protein. Complete proteins are considered higher quality proteins. They come from foods that contain all of the essential amino acids with the correct quantity and ratio to maintain nitrogen balance and allow tissue growth and repair. Incomplete proteins are considered lower-quality proteins because they lack one of more of the essential amino acids [7].
Relevant to bone physiology, the collagenous structural proteins provide scaffolding for minerals and make up sixty percent of bone volume. The collagenous proteins also compose the hair, skin, nails, tendons, and ligaments [7]. Another role that proteins play is in regulating the pH of the body fluids. They are able to buffer the excess acid metabolites that are formed during relatively intense exercise to bring the body back to homeostasis [7].

Relation of Protein to BMD

There are sources of literature that maintain that a diet too high in protein is potentially damaging to bone [1]. The belief behind such literature is that consuming excess protein can lead to increased calcium concentration in the urine and a possibility of increased fracture risk. The thought is that the protein causes an increase in bodily acid production that in turn leaches calcium from the bones to neutralize the upsurge of acid [1]. The loss of calcium from the bones could leave them at a greater risk for fracture, or if not managed, osteoporosis (lower BMD).

Other sources state that consuming ample protein is not detrimental to the overall health of the individual choosing to partake in a high protein diet [15, 16, 17, 18]. In contrast to what some authors believe, that too much protein is harmful, various sources state that the calcium observed in the urine of persons consuming a diet high in protein is excess calcium that the body cannot absorb and thus must be excreted. Both sides of the debate agree that surplus protein leads to calcium in the urine, but where the disagreement lies is why the mineral is appearing in the urine [1, 17, 19].
There have been researchers that concluded additional protein in a diet supports intestinal calcium absorption and decreases the percentage of calcium in the urine that is derived from bone [15]. Thorpe, et al (2008), reported that a diet, which targeted lean protein, sources including low-fat dairy products, preserved bone loss when compared to an isoenergetic diet that was higher in carbohydrates [15].

*Other Bone Related Nutrients*

An interesting phenomenon that researchers have found is that high protein diets are also high in calcium [20]. Calcium is an important mineral for bone formation. Along with calcium, it is well known that high protein diets are also high in phosphorus, vitamin D, and potassium. A diet that has an elevated level of protein appears to additionally contain many needed ingredients for bone health [20]. Optimal BMD can be achieved and maintained on a high protein diet, possibly without exercise, because of the adequate nutrition for bone health that is consumed.
CHAPTER III
METHODOLOGY

Participants

Twenty healthy subjects between 21-38 years of age volunteered to participate in earlier research from which this study’s data will be derived. Each participant has identified himself as a weight trainer for greater than three years. The average amount of time the participants stated that they took in excess protein was nine years.

All participants read and signed consent forms approved by the University of Akron Human Subjects Review Board. Testing procedures, equipment and recording forms were verbally explained to the subjects. Subjects were excluded from participation if they were using Creatine or had any musculoskeletal or metabolic disease. The exclusion limitations were necessary because of a separate part of the study on renal function that required these participants not be included for the accuracy of the data.

For confidentiality purposes, each participant was assigned a number and either letter A or B. Letter A was assigned to participants who attempted to consume ample protein while letter B was given to participants who did not make an effort to add extra protein to their diets. Due to the observational nature of the research design, group assignments were not blinded; participants and most of the researchers, except for the DEXA technician, knew whether a subject was seeking extra dietary protein. The
subjects were asked to use their assigned code on all of their forms and when undergoing any testing for the study.

*Sampling Process*

The sampling process chosen for this research study was convenience sampling. Flyers were posted in multiple locations where individuals in the population would have a greater chance to see them. The main locations that flyers were posted were gyms and fitness centers, as that is the primary location to search for our target population. Participants were also told about the study through word of mouth.

Our sampling method was chosen because of the type of population that our study called for and the necessity to find willing and able volunteers. Our study was completely made of volunteer subjects who met with the researchers to learn about the study before agreeing to participate in the research. A double blind random sampling process would have been ideal for an experimental design, but was not feasible for the current, long duration research study.

*Seven-Day Food Diary*

Each participant was given a seven-day food diary with instruction on how to record his diet for one week. Participants were asked to record anything that they ate or drank. Descriptions for a sample of typical serving sizes of foods were given to participants so that they could accurately record the amount of food consumed. Participants were told to be as accurate with their measurements as they could. This approach to assessing dietary intakes is widely used and valid.
Diet Analyzing

A food-analyzing program, NutriCalc Plus (ESHA Research, Salem, OR), was used to analyze the seven-day food records. Everything the participants recorded on the diet logs was entered into the diet-analyzing program by the same researcher to limit differences in data entry. Through NutriCalc Plus, the researchers were able to see data for individual days as well as a four-day, in-training, average and a seven-day, in training plus non-exercise days, average. The reason for having in-training and rest day diet records was for a separate study using the same data for protein and renal function research.

The researchers chose a four-day average to display the diet of the participant while they were weight training. The last three days of the study, the participants were asked to refrain from weight training. This was done because other data collection that required exercise abstinence was involved and it offered insight into dietary differences on training days versus non-training days.

Dual X-Ray Absorptiometry (DEXA)

Bone Mineral Density was assessed using dual x-ray absorptiometry (DEXA) (Hologic 4500, Bedford, MA). The DEXA was properly calibrated and operated by an experienced radiation technician at Kent State University (Kent, Ohio). Total-body BMD may be confounded by large body size therefore specific anatomical sites were also analyzed in order to address the possible issue.

Fracture risk is measured in standard deviations (SD) by comparing the patient’s bone mass with the bone mass of a healthy, gender-matched twenty five to thirty five-
year old person [4]. Test scores are reported with T- and Z-scores. The T-score represents the number of SDs for the patient compared with normal young adults with mean peak bone mass. The Z-score is the number of SDs for the patient compared with normal person in the same age category [4]. According to the World Health Organization, T-scores greater than 2.5 could confirm a diagnosis of osteoporosis; T-scores between 2.5 and 1.0 are associated with osteopenia; and T-scores less than or equal to 1.0 are considered normal [4].

Statistical Analysis

Data are preset at mean plus-or-minus standard deviation. Group differences were analyzed using unpaired t-tests (Statistica, Statsoft, Inc.). Data were considered significant if it produced a p-value equal to or less than 0.05. From the data, graphs were made to illustrate the results using the same software.
CHAPTER IV

RESULTS

This study was performed to assess the impact of consuming excess protein on bone mineral density specifically on the population of Caucasian male resistance trainers. Twenty resistance trainers, who met study’s criteria, volunteered to participate in the study. There were twelve participants in the protein seeking group and eight participants in the non-protein seeking group. All twenty participants completed the study as asked.

Sample Size

This study had a beginning sample size of twenty-three participants. Three participants had to be excluded from the site-specific statistical analysis due to lack of necessary information regarding site-specific data, leaving a sample size of twenty. This explains the contrasting ‘N’ value in Figure 4.1. If the three participants that were excluded from site-specific bone mineral density data were excluded from that graph, the values did not change.

Demographic Information Regarding Participants

A constraint of this study was that the researchers were unable to have a matched population design for body mass between the two groups. The two groups were not significantly different in terms of age, as can be seen from Figure 4.1. The mean age of the protein-seeking group was 28.46 years old, while the non-protein seeking group had
an average age of 27.6 years old. There was no significant difference between the two groups in the number of years that the participants had been lifting. The protein-seeking group was, however, notably larger than was the non-protein seeking group in terms of body mass.

Figure 4.1. Age of resistance trainers.

Protein Consumption of Participants

The protein-seeking group consumed a significantly larger amount of protein than did the non-protein seeking group (p=0.00009) (Figure 4.2). The protein-seeking group, on average, ate 3.17 grams of protein for every kilogram of body mass compared to the non-protein seeking group consumed 1.5 grams of protein for every kilogram of body mass.
Figure 4.2. Relative protein intake of resistance trainers.

Exercise Information Regarding Participants

A variable in our study that could impact the outcome of the results is exercise. We asked the participants to record their workouts, including exercises (repetitions and sets), amount of time, and rate of perceived exertion (RPE). Based on the data provided, a $t$-test was performed, and there was no significant difference in the RPE of the workouts between the two groups (Figure 4.3).
We also calculated the energy expenditure for the two groups to see if there was a significant difference in the workouts between the groups. Figure 4.4 shows that there is no significant difference in energy expenditure, measured in kcal/kg*week⁻¹, between the protein-seeking group and the non-protein seeking group. To our knowledge, this is the first time that exercise and exertion has been examined in this population with protein and BMD.
Figure 4.4. Energy expenditure of resistance trainers.

Lumbar Spine

We found that the average BMD for the lumbar spine in the protein seekers was 1.16 g/cm² compared to the non-protein seekers who had a lumbar spine BMD of 1.08 g/cm² (Figure 4.4). Based on the data, there was no significant difference in lumbar spine BMD between the two groups (p=0.23). Both groups had a higher BMD than would be seen on an average person (1.05 g/cm²).
We found that the mean BMD for the femoral neck in the protein-seeking group was 1.05 g/cm² whereas the average BMD in the femoral neck region in the non-protein seeking group was 0.96 g/cm² (Figure 4.5). There was not a significant difference between the two groups based on the data collected. The protein seeking group showed a femoral neck BMD similar to the national average where the non-protein seeking group displayed a femoral neck BMD that was lower than ideal based on the data mean plus-or-minus the standard deviation.
Figure 4.6. Femoral neck BMD of resistance trainers.

Summary

In general, the results of this investigation demonstrate significant (p<0.05) differences in protein intake, no difference (p>0.05) in exercise intensity (RPE) or duration (kcal/kg*week^{-1}), and no differences (p>0.05) in the site-specific BMD. The BMD in both the lumbar spine and femoral neck regions were seen to be higher in the protein-seeking group, but the results were not significant.
CHAPTER V
DISCUSSION

Introduction

Diets that are higher in protein have previously been considered unsafe for bone health. In recent years, research has emerged showing that high protein diets are safe and do not harm the integrity of bones [15, 16, 17, 18]. A specific segment of the population, whom are known to consume extra protein, was studied to see the effects of additional protein on site-specific bone health.

The recommended dietary allowance (RDA) for protein is 0.8 grams per kilogram body mass per day [14]. The participants that did not attempt to consume additional protein were above the RDA for protein while the participants that did try to consume extra protein were two to three times, and on occasion higher, than the RDA for protein. The RDA was set to meet the needs of an average, sedentary person, which could be seen as meeting the needs of a majority of the people in the United States.

The problem with those recommendations and athletes, specifically resistance trainers, is that they do not fall into the category of an average, sedentary person. Their protein needs are higher to meet the demands placed on the body by the rigorous activity the athlete participates in [21, 22]. There was a significant (p<0.05) difference in protein intake between the studied groups. The participants that made an effort to add extra
protein to their diet were well above what is recommended and were able to demonstrate the effects of a high protein diet on site-specific bone health.

The International Society of Sports Nutrition (ISSN) concludes that a daily intake of 1.4-2.0 grams per kilogram body mass is not only safe but could improve the training of an athlete [21]. Our non-protein seeking athletes fell on the low end of the ISSN recommendations while our protein-seeking athletes were well above those recommendations. The protein-seekers consumed, on average, 3.1 grams of protein per kilogram of body mass whereas the ISSN suggests up to 2.0 grams of protein per kilogram of body mass [21].

*Lumbar Spine*

Thorpe et al. (2007) found that increasing dietary protein was beneficial to lumbar spine BMD of postmenopausal women [15]. Other studies have also shown an increase in lumbar spine BMD in women that had the highest percentage of calorie intake coming from protein [23, 24]. One study showed that, in men, an increased amount of animal protein did not cause a significant increase in BMD whereas in the same study the increase in BMD was significant for women [23].

Our research appears to correlate with other studies in the literature. Our study showed that protein-seeking resistance trainers did have a higher lumbar spine BMD than did non-protein seeking resistance trainers, but the difference was not significant (p=0.23). Both groups of participants did exhibit a BMD that is considered above average [26] with the protein-seeking group displaying higher numbers than the non-protein seeking group, 1.16 g/cm² and 1.08 g/cm² respectively. We believe these results to be
consistent with what would be seen with a larger sample size. Recruiting a larger population size to test these assumptions would not be feasible for our study.

*Femoral Neck*

As was seen in the lumbar spine, BMD in the femoral neck was shown to be higher in women that consumed a higher percentage of protein [23, 24]. One study found that women who had a low protein intake had a 2.5-3.0% lower hip BMD than those who consumed a moderate to high amount of protein within their diet [25]. The femoral neck and total hip regions appear to correlate based on our data.

We found there to be no significant difference in femoral neck BMD between the protein-seeking group and the non-protein seeking group. The protein-seeking group did demonstrate a higher femoral neck BMD than the non-protein seeking group, which agrees with previous research, but the difference was not significant (p=0.26). The protein-seeking group displayed a femoral neck BMD of 1.05 g/cm², which would be considered an average result [26]. The non-protein seeking group, however, showed a femoral neck BMD of 0.96 g/cm², which would be considered below average [26].

The result that the researchers found to be interesting and unexpected was the lower than normal BMD for the non-protein seeking resistance trainers. It is accepted that resistance athletes have a higher BMD than do non-resistance trainers [10, 11]. It was assumed that site-specific BMD would correlate with total body BMD and be above average in all regions. This assumption was shown to be incorrect as the femoral neck BMD of the non-protein seeking group was below average. One reason this result was
seen could be because the researchers did not look at specific movements the resistance
trainers performed which could possibly play a role in BMD.

Summary

As hypothesized, this research showed that there was no significant difference
between protein-seeking resistance trainers and non-protein seeking resistance trainers in
relation to site-specific BMD. This research also displayed that site-specific BMD can
produce different results than seen with the whole body BMD.

Further research could be done concerning protein type, such as whey, soy, egg,
meat, and BMD for this population. One also might consider researching effects of
protein on BMD in minority resistance trainers, i.e., African Americans, those of Asian
descent, or the Hispanic population. One thing that might have improved this study is an
extended diet record to gain a better idea of average dietary consumption beyond just
protein. For example, future research should examine a combination of dietary and
urinary calcium content and its interaction with protein and resistance training.
REFERENCES


