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**Serum concentration levels of 25(OH)D and injury reports in NCAA Division I football players.**

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# Serum concentration levels of 25(OH)D and injury reports in NCAA Division I football players.

by

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**Report**

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**Serum concentration levels of 25(OH)D and injury reports in NCAA Division I football players.**

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Vitamin D deficiency has been linked with many health problems. Early research demonstrated the importance of vitamin D for bone health, but it may also play a larger role than first reported in muscle health and function. Specifically, low vitamin D may hinder athletic performance, as such evaluation of serum vitamin D levels in high volume training athletes has merit. The purpose of this study was to evaluate serum levels of 25(OH)D in college athletes to determine how many had levels below the recommended values. Data from student-athletes who were attending a large university in the south included: serum vitamin D levels, demographics information, and injury reports. Mean serum vitamin D level for the group was 34.17 ng/mL ± 0.88. Average injury for the group was 1.3± 0.14. The mean value of serum vitamin D for Caucasian players was 38.3 ng/mL ± 1.33 with a range of 23-59 ng/mL. The mean value of serum vitamin D for African American players was 31.16 ng/mL ± 1.08 with a range of 16-52 ng/mL. African American players had significantly lower serum vitamin D levels ($p<0.01$) than Caucasian players. Players with one or more injury had significantly lower serum vitamin D values ($p<0.05$) than players who had zero injuries. Forty-eight players (44.4%) had insufficient levels of vitamin D (20-31.9ng/ml). 60 players (55.6%) had values within normal limits (>32 ng/ml). Players with one or more musculoskeletal injury or fracture had significantly lower serum vitamin D levels ($p<0.05$) compared to players that had zero injuries. African American players had significantly lower serum vitamin D levels ($p<0.01$) compared to Caucasian players. It is important for athletes to monitor serum vitamin D levels and adhere to a supplementation protocol when levels are insufficient.

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## Introduction

Vitamin D is necessary for bone health, calcium absorption, and strengthens the immune system, while deficiency can lead to many poor health outcomes including: reduced immunity, increased inflammation, and poor muscle function. Therefore, maintaining adequate levels of vitamin D may reduce stress fractures, total body inflammation, common infectious illnesses, and impaired muscle function. A healthy level of serum 25(OH)D concentration is considered greater than 30 ng/ml, with optimal levels being greater than 40 ng/ml (Larson-Meyer, 2013). Basic vitamin D functions are reached at greater than 32 ng/mL, but only at levels higher than 40 ng/mL does vitamin D begin to be stored in fat and muscle for future use (Cannell, Hollis, Sorenson, Taft & Anderson, 2009). Serum levels of 40 ng/mL are needed for optimal fracture prevention, and levels of 50 ng/mL are needed for peak performance in elite athletes (Shuler, Wingate, Moore & Giangarra, 2012). Health enhancing levels of vitamin D can largely be achieved in two ways: (a) consumption and (b) ultraviolet rays.

Under the right conditions human can readily produce sufficient vitamin D; however, dietary choices and skin pigment are among the emerging inhibitors of having adequate levels of serum vitamin D. Vitamin D deficiency is related to reduced skin synthesis, or reduced dietary intake and absorption (Angeline, Gee, Shindle, Warren & Rodeo, 2013). The majority of vitamin D is obtained through the sunlight. Athletes that practice indoors, and athletes with darker skin pigmentation are affected by reduced skin synthesis of vitamin D. Melanin absorbs UVB light, reducing vitamin D synthesis; the more melanin, the darker the skin, and the longer the sun exposure needed to produce vitamin D (Angeline et al., 2013).

Vitamin D deficiency causes immediate risks including stress fractures. It is still unknown whether Vitamin D deficiency may cause immediate risk to muscle, tendon and ligament tissue (Hamilton, 2011). The discovery of the Vitamin D Receptor (VDR) in skeletal muscle tissue is good evidence that vitamin D deficiency may have a direct role in skeletal muscle health (Hamilton, 2011). Muscle biopsies in adults with vitamin D deficiency have revealed type II muscle atrophy  (Ceglia, 2008). Type II muscle fibers are involved in power movements. Football is a very explosive sport, and type II muscle fiber atrophy could hinder performance.

Adequate vitamin D levels are critical to overall health and performance of athletes, and this is why it is of interest to explore what optimal serum levels are, a proper supplementation protocol, and consequences vitamin D deficiency. Accordingly, the purpose of this study was to evaluate the serum concentration of 25(OH)D in collegiate football players during the season to determine whether vitamin D levels were below or above recommended values. Moreover, this study would compare the levels of serum vitamin D with recent injuries to examine the association of the two. When comparing groups with similar serum vitamin D levels it is hypothesized that players with lower serum vitamin D levels will have more fractures and musculoskeletal injuries.

## Methods

After IRB approval was granted, injury reports and serum vitamin D levels were collected from 108 NCAA Division I football players were provided to the researchers for analysis. All of the participants were between 18-22 years of age ($M = 20.00 \pm 1.15$), with 53% of the athletes being African American, 41% Caucasian, and 6% of another race.

### Procedures

Prior to the start of the competitive football season, but after pre-season workouts, venipunctures were performed on the athletes by a professional, clinical pathologies lab located in the same state where the football program resided. Assays were conducted by the laboratory to determine the serum 25(OH)D concentrations within each blood sample.

A volunteer unaffiliated with this research project created a spreadsheet of data with subject IDs to replace the athletes' names. Data included in the spreadsheet that was provided to the researcher were: (a) vitamin D concentration from September lab draw, (b) age, (c) ethnicity, (d) vitamin D supplementation history, (d) position, and (e) total injuries (fracture and musculoskeletal) within three months prior to laboratory draw and two months post laboratory draw.

### Data Analysis

Descriptive statistics and analysis of variance were used to analyze the data. These data were presented as means ± SE. Mean vitamin D differences between players with zero injuries and one or more injury were compared using an unpaired Student's t-Test. Mean vitamin D differences between Caucasian and African American players were compared using an unpaired Student's t-Test. An alpha level of $p < .05$ was set for all analyses in this study

## Results

Mean serum vitamin D level for the group was 34.17 ng/mL ± 0.88. Average injury for the group was 1.3± 0.14. The mean value of serum vitamin D for Caucasian players was 38.3 ng/mL ± 1.35 with a range of 23-59 ng/mL. The mean value of serum vitamin D for African American players was 31.16 ng/mL ± 1.08 with a range of 16-52 ng/mL. 48 players (44.4%) had insufficient levels of vitamin D (20-31.9ng/ml). Sixty players (55.6%) had values within normal limits (>32 ng/ml). African American players (M = 31.16; SE = 1.08) had significantly lower serum vitamin D levels ($p<0.01$) than Caucasian players (M = 38.3; SE = 1.33). Players with one or more injuries had significantly lower serum vitamin D values ($p<0.05$) than players who had zero injuries.

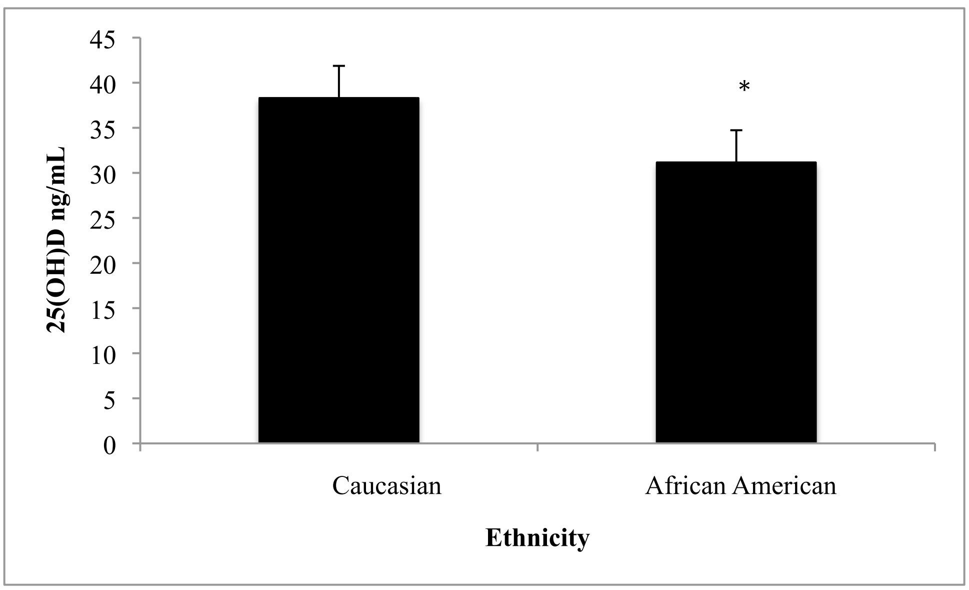


**Figure 1.** *Mean serum 25(OH)D ng/mL in Caucasian players and African American players. Bars represent means ± SE. Serum levels significantly lower in African American players compared to Caucasian players.*

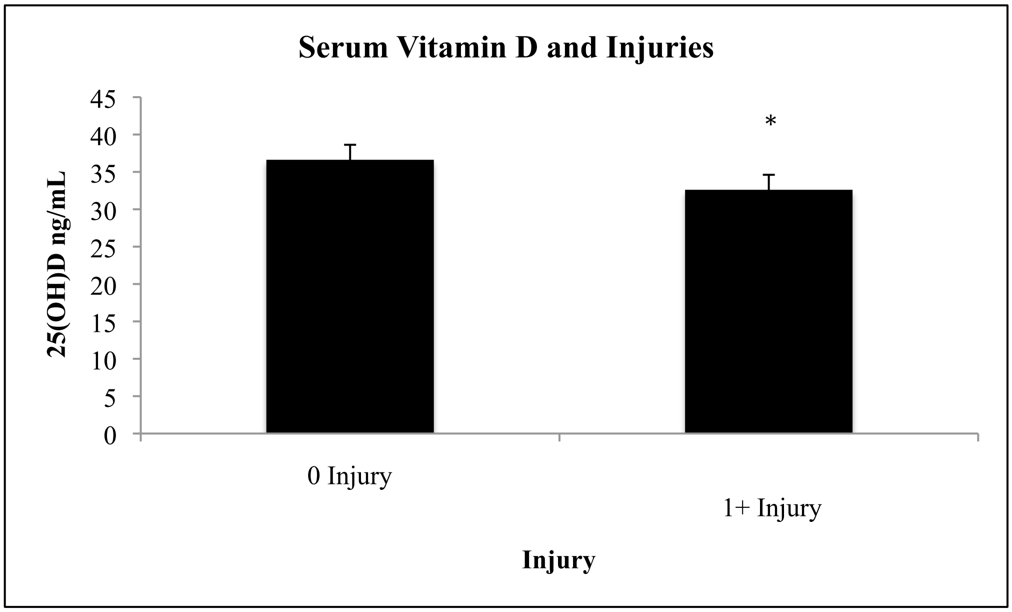


**Figure 2.** *Mean serum 25(OH)D ng/mL in players without injury, and players with one or more injury. Bars represent means ± SE. Serum levels significantly lower in players with one or more injuries.*

## Discussion

Vitamin D plays a significant role in bone health, calcium absorption, and in the immune system, yet many division athletes fail to maintain minimal levels, which can prevent injury. Several significant findings may be influential to the training and diet of such athletes. In this present study, African American players had significantly lower serum vitamin D levels ($p<0.01$) than Caucasian players. These findings may be explained by the difference in skin pigment (Peeling, Fulton, Binnie & Goodman, 2013). Darker skinned humans have a difficult time in creating vitamin D through ultraviolet light. This is because the more pigment in the skin makes it harder for the UVB rays to penetrate past surface level.

Typical diets do not contain high levels of vitamin D unless large consumption of wild caught fatty fish is included (Cannell et al., 2009). Most of the vitamin D obtained is from exposure to sunlight. Melanin competes for UVB rays, and the more melanin in the skin, the longer the exposure to UVB rays need to synthesize vitamin D (Angeline et al., 2013). African American athletes had significantly lower serum vitamin D levels compared to their Caucasian teammates. The amount of pigmentation in skin was not determined, and the level of melanin could have varied greatly within groups. If players could have been divided by pigmentation rather than ethnicity, greater differences may have been observed. Further studies should divide groups by pigmentation when comparing serum vitamin D values.

### Injuries and Vitamin D

In this study, players with one or more musculoskeletal injuries or fractures had significantly lower serum vitamin D levels compared to players that had zero musculoskeletal or fracture injury. Injury reports were three months pre- or post-blood draw, and included musculoskeletal injuries and fractures.

Players that reported musculoskeletal injuries and fractures during the season had significantly lower serum 25(OH)D levels than players that did not obtain injuries. This is of particular interest for the medical staff, and reason to monitor serum vitamin D levels in athletes closely throughout the season, and supplement if levels are not sufficient. It is known that vitamin D regulates growth signal transduction pathways in skeletal muscles, and vitamin D deficiency leads to muscle weakness (Christakos et al., 2013). Decreased vitamin D is also a well-known risk for stress fracture because of the parathyroid hormone activity that regulates calcium is increased, pulling calcium from bones (Ogan & Pritchett, 2013).

Despite the fact that vitamin D supplementation was provided for athletes with low serum levels throughout the season, vitamin D deficiencies still existed. Compliance varied, but common regimens were 1000-2000 IU, 2-3 times a week. According to the study by Chao et al., the minimum dose of 1000-2000 IU, did not significantly increase serum vitamin D levels unless supplementation was provided for 3-5 months (2013). Since the injury reports provided were three months pre-blood draw and two months post-blood draw, it can be assumed that extreme fluctuation in serum levels did not occur. Future research should investigate follow-up laboratory values after supplementation.

The autocrine pathway uses vitamin D to regulate immune function, inflammation response, cell turnover, protein synthesis, hormone synthesis, and gene expression (Heaney, 2008). Lack of vitamin D leads to a suppressed immune system and increased inflammation. Reduced vitamin D levels may down-regulate the anti-microbial peptides, involved in innate immunity, leading to vitamin D deficient athletes having depressed immune function (Peeling et al., 2013). It would be of interest in future studies to track illnesses throughout the season and compare vitamin D levels to see if there is a significant relationship. Athletes continually stimulate the inflammatory response through the stress of exercise. Having adequate vitamin D levels may reduce inflammation, leading to better performance. Exploring inflammatory markers and vitamin D levels would be a positive addition to future research.

Vitamin D deficiency has been linked to type II muscle fiber atrophy (Cannell, 2009). This has severe ramifications for a high-power sport such as football. 44% of the players had insufficient levels of vitamin D. Levels below 32 ng/mL hinder genomic and non-genomic effects of vitamin D. Optimal calcium absorption requires levels of greater than 30 ng/mL, and only at levels greater than 40 ng/mL does fracture prevention occur (Shuler et al., 2012). Vitamin D has recently been discovered to impact protein synthesis through the VDR (Ogan & Pritchett, 2013). It is essential to monitor athletes' serum vitamin D levels, especially during peak training times. If levels are not sufficient it is important to supplement until optimal levels are reached in order to prevent injuries, and promote optimal performance.

## Limitations

Only one laboratory blood draw was provided during the season. It would be beneficial to obtain post-season laboratory values in order to determine the fluctuations of the serum vitamin D levels in the athletes. Compliance with vitamin D supplementation was also not given, which would have been helpful if pre- and post-season laboratory values were taken in order to determine the effectiveness of the supplementation protocol. Ethnicity was determined by the player handbook, and does not accurately reflect varying pigmentation.

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## Literature Review

### Introduction

Vitamin D deficiency can lead to many poor health outcomes including: reduced immunity, increased inflammation, poor muscle function, and a negative effect on bone health. Therefore, maintaining adequate levels of vitamin D may reduce stress fractures, total body inflammation, common infectious illnesses, and impaired muscle function. A healthy level of serum 25(OH)D concentration is considered greater than 30 ng/ml, with optimal levels being greater than 40 ng/ml (Larson-Meyer, 2013). Basic vitamin D functions are reached at greater than 32 ng/mL, but only at levels higher than 40 ng/mL does vitamin D begin to be stored in fat and muscle for future use (Cannell, Hollis, Sorenson, Taft & Anderson, 2009). Serum levels of 40 ng/mL are needed for optimal fracture prevention, and levels of 50ng/mL are needed for peak performance in elite athletes (Shuler, Wingate, Moore & Giangarra, 2012).

Adequate vitamin D levels are critical to overall health and performance of athletes, and this is why it is of interest to explore what optimal serum levels are, a proper supplementation protocol, and consequences of vitamin D deficiency.

### Physiology

Vitamin D has functions in both the endocrine and autocrine systems. Vitamin D enhances intestinal calcium absorption and osteoclast activity, which is why vitamin D is essential for bone growth. With lower levels of vitamin D, parathyroid hormone increases to regulate the body’s demand for calcium, leading to bone releasing more calcium (Ogan & Pritchett, 2013). Low levels of vitamin D increase parathyroid hormone activity, which increases risk for bone injury. For this reason, it is important to monitor the serum vitamin D levels in athletes to avoid insufficiency.

The majority of the vitamin D in the body is used by autocrine mechanisms on a daily basis (Heaney, 2008). These varying intracellular mechanisms regulate gene expression. Vitamin D Receptors (VDR) have been discovered in almost every tissue of the body, leading to the belief that vitamin D has a much larger role in human physiology than previously believed (Ogan & Pritchett, 2013). The autocrine pathway uses vitamin D to regulate immune function, inflammation response, cell turnover, protein synthesis, hormone synthesis, and gene expression (Heaney, 2008). The discovery of the vitamin D Receptor in muscle suggests the importance of vitamin D in muscle function (Ogan & Pritchett, 2013).

Vitamin D is a potent antioxidant that protects against free radical damage. Vitamin D may induce cell differentiation, leading to anticancer activity (Angeline, Gee, Shindle, Warren & Rodeo, 2013). The VDR is affects muscle strength and stability. Vitamin D acts as a hormone on nuclear receptors that control cellular functions including: protein synthesis, mRNA transcription, increased calcium uptake, and differentiation into muscle fibers (Angeline et al., 2013).

The vitamin D Receptor in muscle is still being studied, but the preliminary findings suggest several mechanisms of vitamin D's regulation of muscle function. One indirect mechanism of muscle function is vitamin D helps regulate the transportation of calcium in the sarcoplasmic reticulum by increasing the efficiency, or number of calcium binding sites during muscle contraction (Ogan & Pritchett, 2013). Vitamin D also regulates protein synthesis through autocrine mechanisms, making vitamin D essential for optimal muscle function (Ogan & Pritchett, 2013). Myopathy is a common symptom of vitamin D deficiency. Alterations in intracellular calcium ion regulation involved in muscle contraction due to lower levels in vitamin

D may account for the skeletal muscle weakness described as a symptom of vitamin D deficiency. (Christakos et al., 2013).

## Vitamin D and Muscle

The VDR found in skeletal muscle is evidence that vitamin D has a direct role in muscle function. Muscle biopsies in adults with vitamin D deficiency have revealed type II muscle atrophy  (Ceglia, 2008). Type II muscle fibers are involved in power movements. Football is a very explosive sport, and type II muscle fiber atrophy could hinder performance. Vitamin D deficiency causes immediate risks including stress fractures. It is still unknown whether vitamin D deficiency may cause immediate risk to muscle, tendon and ligament tissue (Hamilton, 2011).

The discovery of the VDR in skeletal muscle tissue is good evidence that vitamin D deficiency may have a direct role in skeletal muscle health (Hamilton, 2011). A Vitamin D Response Element (VDRE) has been identified in the promoter region for human Insulin-like growth factor binding protein 3 (IGFBP-3). Vitamin D is believed to have a regulatory effect on IGFBP-3 because of this VDRE. The VDR binds to and regulates the expression of certain genes. This is significant in regards to IGF-1 function because IGF-1 induces proliferation, differentiation and hypertrophy (Hamilton, 2011). The discovery of the VDRE in the promoter of IGFBP-3 provides evidence that vitamin D has an important role of skeletal muscle function because the binding of IGFBP-3 to IGF-1 can inhibit or stimulate IGF-1's function (Hamilton, 2011).

## Vitamin D Synthesis

Vitamin D deficiency is related to reduced skin synthesis, or reduced dietary intake and absorption (Angeline et al., 2013). The majority of vitamin D is obtained through the sunlight. UVB radiation converts 7-dehydrocholesterol to pre-vitamin $D_3$, which is then converted to cholecalciferol (vitamin $D_3$) in the skin (Moran, McClung, Khoen & Leiberman, 2013). Athletes that practice indoors, and athletes with darker skin pigmentation are affected by reduced skin synthesis of vitamin D. Melanin absorbs UVB light, reducing vitamin D synthesis. The more melanin, the darker the skin, and the longer the sun exposure needed to produce vitamin D (Angeline et al., 2013).

Chen et al., studied the effects of UVB radiation on different skin pigmentation to determine the corresponding circulating vitamin D levels (2007). After the same amount of UVB exposure, the black subjects had little detectable vitamin $D_3$ circulating in the blood compared to the white control subjects. The threshold of UVB exposure needed to synthesize previtamin $D_3$ was higher in blacks than whites (Chen et al., 2007).

Clothing and exposure to sunlight pose risks to vitamin D synthesis. In athletic populations it is important to consider the uniforms and training facilities when monitoring vitamin D levels. Sun block and excessive clothing block the absorption of UVB rays, leading to less synthesis of vitamin D (Webb, 2006). Latitude, season and time of day are all factors in levels of UVB absorption. Certain months of the year when the rays are not as intense, vitamin D supplementation may be necessary. Personal factors, such as clothing worn, sunscreen use, and skin pigmentation are all factors effecting vitamin D synthesis (Webb, 2006).

**Vitamin D Recommendations**

Sources vary when determining the proper amount of serum vitamin D levels and supplementation recommendations. Chao et al., evaluated the relationship between dose, frequency and duration of supplementation to increase plasma 25-Hydroxyvitamin 2 (2013). The minimum dose tested was 1000-2000 IU once or twice per week for a month, and this regimen was not found to increase 25(OH)D levels (Chao et al., 2013). Higher doses, 2000-5000 IU, three to seven times per week, for five months or longer, increased serum vitamin D levels. Dose and frequency had more pronounced effects of increasing serum vitamin D levels, and duration only significantly increased serum vitamin D levels when supplementation occurred three to five months (Chao et al., 2013).

The endocrine society recommends an intake of 1500-2000 IU per day for adults ages 19-70 years old (Ogan & Pritchett, 2013). Parathyroid hormone levels become stable at 25(OH)D levels greater than 32 ng/mL, lower levels of vitamin D may result in hyperparathyroidism. Hyperparathyroidism may lead to muscle and bone problems (Ogan & Pritchett, 2013). 25(OH)D levels above 40 ng/mL result in vitamin D being stored in the muscle and fat.  Levels below 40 ng/mL are sufficient to meet the daily metabolic needs. 3000-5000 IU of vitamin D per day is recommended to meet the needs of every tissue in the body (Ogan & Pritchett, 2013).

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