# The relationship between vitamin D levels, injury and muscle function in adolescent dancers.

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#### Abstract

Vitamin D has been shown to benefit a diverse range of health functions including muscle function. The aim of the present study was to identify serum 25(OH)D₃ levels in a sample of adolescent dancers and compare them to muscle function and injury incidence. We incorporated a cross-sectional design to study 49 pre-professional male and female dancers (17±4.44yrs, 52.1±6.72kg, 1.63±0.07m) in full-time training in Brazil. Serum 25[OH]D<sub>3</sub> was analyzed by Enzyme-linked Immunosorbent Assay; quadriceps and hamstring peak torque and muscle fatigue were measured by isokinetic dynamometer at 60 and 300° s<sup>-1</sup>. Injury type and location in the previous 6months were determined by self-report questionnaire. Participants were categorized into 2 groups: normal or insufficient/deficient (>or<30 ng/ml 25[OH]D<sub>3</sub>). Results indicated the normal serum 25[OH]D₃ group had significantly lower fatigue rates than the insufficient/deficient group (p<0.05) but not for the other muscle function parameters. Fifty-seven percent of participants reported at least 1 injury. The most common were sprains (33%) and tendinopathies (19%). Injured dancers had significantly lower peak torque at 60°/s. The link between serum 25[OH]D₃ and reduced muscle fatigue resistance has not been shown before, though the underlying mechanisms aren't apparent and the link between muscular strength and injury has been previously evidenced.

Keywords: Muscle Strength Dynamometer, fatigue, dance, serum 25(OH)D.

#### **INTRODUCTION**

Pre-professional dance students undertake 3-6 hours of dance training a day; this is generally technique focused but at different times of the year will also include rehearsals and performances [1, 2]. The training regimen is often perceived as exhaustive and intense leading to several health problems and a high injury prevalence [3, 4]. The injury prevalence has been attributed to the long training hours the high relative intensity, the impact from jumping and muscular imbalances [5, 6]. A lean and ectomorphic body is required for classical ballet professionals[7] which has been linked to poor nutritional practices[8].

Vitamin D has several effects on body systems[9] and has an influence on muscle function[10]. Vitamin D is linked to metabolism and protein synthesis in skeletal muscle cells[11]. Dancers have been shown to be vitamin D insufficient/deficient throughout the year, with only slight improvements during the summer months, potentially due to their long hours training indoors[12]. Supplementation has been shown to improve vitamin D levels with subsequent improvements in muscle function and a reduction in injury incidence[13, 14].

There are still few studies relating to classical ballet dancing and vitamin D levels in warm climate countries like Brazil, although studies on the general population in high sunlight countries have indicated similar deficiency incidence[15]. Considering that dancers are often involved in technical classes/rehearsals indoors for several hours a day and that such activities make them more susceptible to musculoskeletal injuries, we hypothesize that, in dancers, insufficient levels of vitamin D are related to a decrease in muscle function and a higher injury incidence. Therefore, the aim was to identify serum 25[OH]D<sub>3</sub>, muscle function and injury incidence in adolescent vocational ballet dancers.

#### **MATERIALS AND METHODS**

The study, a cross-sectional design, was approved by the lead author's university Ethics and Research Committee[16]. Data collection took place in Goiania at latitude - 16.6799, longitude -49.255 during the month of October; the previous three months mean sunlight was 11.5 hours (http://www.inmet.gov.br/). The inclusion criteria were as follows: male and female dancers, aged 13 years and older, who have been dancing for more than 5 years and who were duly enrolled in Centro de Educação Profissional em Artes Basileu França (CEPABF). Dancers who had been absent from training for 1 month, were under medical care or injured and younger than 13 years old were excluded. Fifty-eight dancers were initially recruited, 49 dancers presented for blood collection and a further 7 did not attend the muscle strength evaluation; therefore, the final sample of the present study was composed of 42 dancers (Table 1). All participants with the exception of two were white Caucasians.

Table 1: Participant information

|                | Age<br>(yrs) | Body Mass<br>(kg) | Stature<br>(m) | ВМІ        | Age<br>started<br>dancing<br>(yrs) | Years<br>dancing<br>(yrs) | Dance<br>exposure<br>per week<br>(hrs) |
|----------------|--------------|-------------------|----------------|------------|------------------------------------|---------------------------|--|
| Males (n=5)    | 17 ±4.01     | 61.6 ±7.92        | 1.74 ±0.09     | 20.4 ±0.76 | 15 ±4.85                           | 7 ±3.27                   | 5 ±2.06                                |
| Females (n=37) | 17 ±5.55     | 51.2 ±5.99        | 1.62 ±0.06     | 19.4 ±1.91 | 7 ±2.90                            | 10 ±3.29                  | 5 ±2.02                                |

#### **Procedures**

The dancers' anthropometric and training history data were collected including age, sex, stature, body mass, age they started dancing and daily duration of training. Stature (cm) was measured to the nearest 0.1cm using a SECA 217 stadiometer (Germany) and body mass (kg) to the nearest 0.1kg with SECA 761 scales (Germany) wearing minimal clothing. Age and age they started training were documented as whole years. Daily training was recorded in 30 minute periods. A questionnaire also included questions regarding supplementation consumption (minerals or vitamins) and whether any of the females were taking contraceptive mediciation.

Participant's vitamin D levels were measured via blood sample. The blood collection was performed by ulnar venipuncture, in accordance with the recommendations of the Brazilian Society of Clinical Pathology. A 5-7 ml sample of whole blood was collected into tubes which did not contain anticoagulants. The blood was left to clot for 30 min at room temperature before being spun in a centrifuge at  $1500 \times g$  for 10 min. A 0.5 ml of serum was aliquoted into 2 ml micro-tubes prior to be being stored at -80 °C. The thawed samples were later analyzed for serum  $25(OH)D_3$  by using the enzyme-linked immunosorbent assay (ELISA), through the 25-OH Vitamin D® kit (DRG, Springfield, NJ, USA), with interassay CV of 2.9%. All the collected blood material was analyzed in the Laboratory of Molecular Immunology at UEG Campus Laranjeiras.

Muscle function was evaluated by isokinetic dynamometry of the knee joint using a Biodex System 4 PRO isokinetic dynamometer (Biodex Medical Systems, Shirley, NY, USA). The isokinetic dynamometer was set up with 5° recline of the chairback from the vertical position and the participant positioned on the chair so that the popliteal fossa of the knee was 5 cm away from the chair edge and the arm was positioned and secured by a belt 5 mm above the lateral malleolus. The range of motion was limited to between 10° and 110° of knee flexion. The participant was stabilized by 4 belts; two diagonals to the thorax, one in the lower abdominal region (in the transverse axis) and one in the lateral femoral condyle region of the femur bone of the evaluated limb (on the transverse axis). Participants carried out a moderate intensity cycle ergometer warm up for 5 minutes prior to a series of self-directed dynamic stretches. The test consisted of 3 sets of 5 repetitions for knee concentric flexion/extension at angular velocities of 60°s -1 and 300°s -1 (12,13); the velocity order was randomly assigned for each participant using a sealed envelope method.

Injury history was assessed by self-report questionnaire. Participants were asked to recall injuries over the last 6-months that included anatomical location, injury type (dislocation or subluxation, sprain, fracture, contusion and tendinopathy) and time off from dance activities.

## **Data Analysis**

Vitamin D insufficiency was determined by circulating serum  $25(OH)D_3$  levels of 10-30 ng/ml (25-75 nmol/l) and deficiency as a level below 10 ng/ml (25 nmol/l)[17], participants were grouped according to having either normal levels (>30 ng/ml) or insufficient/deficient levels (<30 ng/ml). Dancers were further grouped according to their injury recall status into either injury or no injury. Fatigue index was calculated from the isokinetic data using the individual's greatest and lowest score over the 15 repetitions for both flexion (hamstrings) and extension (quadriceps). Peak torque was recorded from the isokinetic dynamometer.

# Statistical analysis

Descriptive statistics were calculated for all the muscle function and injury data. Multiple Analysis of Variance tests were used to examine the effect of sex, vitamin D group, movement (extension/flexion), leg (left/right) on peak torque and fatigue index. Skin tone was not used as a covariate as only two participants were not white Caucasians. Chi-square tests were used to examine injury data between normal and insufficient/deficient vitamin D groups and vitamin D levels, and muscle function between injury and no injury groups. The Statistical Package for Social Sciences (SPSS) 23.0 was used for all analyses and significance set at p≤0.05.

## **RESULTS**

None of the participants reported taking mineral or vitamin supplementation and no females reported taking contraceptive medication. Main effects were observed for Sex (F=5.673, p<0.001), Leg (F=9.997, p<0.001), Movement (F=46.18, p<0.001), Sex\*Vitamin D group (F=4.989, p<0.001), Leg\* Movement (F=9.896, p<0.001).

Table 2: Descriptive data (mean $\pm$ sd) for serum 25(OH)D $_3$ , peak torque and fatigue index according to gender, vitamin D group and leg

|                        | Normal       |              |              |              | Insufficient/Deficient |              |             |              |
|------------------------|--------------|--------------|--------------|--------------|------------------------|--------------|-------------|--------------|
|                        | Ma           | ale          | Female       |              | Male                   |              | Female      |              |
| Serum 25(OH)D₃ (ng/ml) | 48.8 ±7.49   |              | 43.3 ±8.74   |              | 5.9 ±0.66              |              | 17.8 ±7.92  |              |
|                        |              |              | ,            |              |                        |              | ,           |              |
| Leg                    | Right        | Left         | Right        | Left         | Right                  | Left         | Right       | Left         |
| Peak Torque            |              |              |              |              |                        |              |             |              |
| 60°.s <sup>-1</sup>    |              |              |              |              |                        |              |             |              |
| Extension (n.m)        | 124.2 ±46.67 | 136.3 ±49.24 | 130.1 ±21.92 | 122.3 ±25.79 | 159.2 ±16.62           | 138.8 ±37.83 | 134. ±34.83 | 119.4 ±36.47 |
| Flexion (n.m)          | 72.3 ±14.31  | 70.4 ±46.39  | 56.6 ±13.37  | 57.5 ±14.31  | 65.7 ±10.75            | 69.4 ±8.42   | 62.9 ±25.76 | 58.3 ±20.68  |
| 300° s <sup>-1</sup>   |              |              |              |              |                        |              |             |              |
| Extension (n.m)        | 86.6 ±45.11  | 86.4 ±39.69  | 69.4 ±11.39  | 66.5 ±12.74  | 80.1 ±13.72            | 65.3 ±21.85  | 64.9 ±19.56 | 63.0 ±19.68  |
| Flexion (n.m)          | 59.4 ±31.79  | 59.0 ±33.13  | 47.3 ±9.28   | 48.3 ±9.87   | 59.4 ±31.79            | 54.5 ±4.38   | 46.1 ±14.89 | 47.6 ±15.74  |
| Fatigue index          |              |              |              |              |                        |              |             |              |
| 60° s <sup>-1</sup>    |              |              |              |              |                        |              |             |              |
| Extension (%)          | 13.3 ±3.16   | 18.9 ±10.54  | 20.3 ±11.09  | 21.2 ±11.93  | 14.6 ±15.49            | -6.9 ±52.12  | 17.6 ±5.40  | 19.4 ±13.89  |
| Flexion (%)            | 10.7 ±15.62  | 18.7 ±5.75   | 16.5 ±15.49  | 19.5 ±10.28  | -16.4 ±40.31           | 10.9 ±20.22  | 3.9 ±27.75  | 18.0 ±8.13   |
| 300° s <sup>-1</sup>   |              |              |              |              |                        |              |             |              |
| Extension (%)          | 22.0 ±7.16   | 25.7 ±12.54  | 27.6 ±13.69  | 28.7 ±11.99  | 10.3 ±42.85            | -0.7 ±52.04  | 19.5 ±16.98 | 21.0 ±15.64  |
| Flexion (%)            | 28.7 ±12.78  | 68.0 ±7.25   | 27.9 ±14.17  | 68.4 ±9.77   | 9.7 ±56.56             | 69.9 ±4.67   | 8.8 ±26.88  | 39.7 ±37.2   |

## Vitamin D

According to the parameters adopted by the Endocrine Society, 26 dancers were classified as having normal  $25(OH)D_3$  levels  $(43.5 \pm 9.09 \text{ ng/ml})$ ; 4 were insufficient  $(25.4 \pm 2.79 \text{ ng/ml})$  and 12 were deficient  $(12.2 \pm 6.14 \text{ ng/ml})$ ; these were subsequently put into 2 groups, normal and insufficient/deficient (Table 2). Between-subject effects reported a Sex\*Group interaction  $(F_{1,152}=17.653; p<0.001)$  with males in the insufficient/deficient group having significantly lower serum  $25(OH)D_3$  than females; there was no difference between the sexes in the normal group. There were significant between-subject effects for the vitamin D groups for fatigue index at  $60^{\circ}s^{-1}$  ( $F_{1,152}=7.319$ ; p<0.01) and  $300^{\circ}s^{-1}$  ( $F_{1,152}=7.440$ ; p<0.01); in both instances the fatigue index was greater in the normal vitamin D group.

#### Muscle function

Irrespective of vitamin D group the following interactions were observed. Between-subject sex effects reported significantly greater extension than flexion peak torques at  $60^{\circ}\text{s}^{-1}$  ( $F_{1,152}$ =119.991; p<0.001) and  $300^{\circ}\text{s}^{-1}$  ( $F_{1,152}$ =27.201; p<0.001). A sex effect was also noted for peak torque at  $300^{\circ}\text{s}^{-1}$  with males having significantly higher scores ( $F_{1,152}$ =6.334; p<0.05). Females reported a significantly higher fatigue index at  $60^{\circ}\text{s}^{-1}$  ( $F_{1,152}$ =9.058; p<0.01) than their male counterparts. The left leg also had a significantly higher fatigue index than the right leg ( $F_{1,152}$ =39.375; p<0.001). Flexion fatigue index was significantly greater than extension data at  $300^{\circ}\text{s}^{-1}$  ( $F_{1,152}$ =38.479; p<0.001)

Table 3: Incidence of injury types for gender, vitamin D group and anatomical site

| Vit D status  |        | Dislocation | Sprain | Fracture* | Contusion* | Tendonitis* | Total |
|---------------|--------|-------------|--------|-----------|------------|-------------|-------|
| Normal        | Male   | 0           | 0      | 1         | 0          | 0           | 1     |
|               | Female | 1           | 8      | 3         | 0          | 5           | 17    |
| Insufficient/ | Male   | 1           | 1      | 0         | 0          | 0           | 2     |
| Deficient     | Female | 4           | 5      | 4         | 3          | 4           | 20    |
|               | Total  | 6           | 14     | 8         | 3          | 9           | 40    |
| Site          |        |             |        |           |            |             |       |
|               | Toe    |             | 2      |           |            |             | 2     |
|               | Foot   | 3           | 4      | 2         |            | 1           | 9     |
|               | Ankle  |             | 2      |           |            | 3           | 5     |
|               | Calf   |             |        |           | 1          |             | 1     |
|               | Knee   | 1           | 3      |           | 1          | 3           | 8     |
|               | Thigh  | 1           |        |           |            |             | 1     |
|               | Leg    |             | 1      |           |            |             | 1     |
|               | Groin  |             |        |           |            | 1           | 1     |
|               | Coccyx | 1           |        |           |            |             | 1     |
|               | Hand   |             |        | 1         |            |             | 1     |
|               | Finger |             | 2      | 1         |            |             | 3     |
|               | Face   |             |        | 1         |            |             | 1     |

<sup>\*</sup>missing data for site

## Injury

Fifty-seven percent of participants reported an injury during the study period. The most common types were sprain (33%), tendinopathy (19%) and fracture (12%) (Table 3) with the majority of injuries were to the lower limbs. Neither sex nor vitamin D status had a significant effect on injury incidence. Injured dancers had significantly lower extension and flexion peak torque at 60°s -1 (p<0.05).

## **DISCUSSION**

The present study aimed to identify levels of vitamin D<sub>3</sub> and to verify the impact of these levels on the parameters of thigh muscle strength and injury status of adolescent classic ballet dance professionals. The data collection occurred during the dry season in Goiânia, Brazil that begins in April and extends until the first half of October that is considered favorable for vitamin D production through solar radiation[18]. Despite the potential solar benefit 43% of the dancers were observed to be insufficient or deficient in serum 25(OH)D<sub>3</sub>; this could be due to the length of time spent indoors[14], the use of sunscreen[19] and a diet poor in vitamin D sources[15]. This is similar to the data reported by Wolman et al[12] that the potential of summer sun exposure only has a negligible effect on dancers' serum 25(OH)D<sub>3</sub> as they are indoors between 10am to 6pm and also use sunscreen. The increased prevalence of deficiency in males was also observed by Looker et al[20], though they could not provide any rationale for the sex difference.

Unlike a number of previous studies on dancers[12-14, 21, 22] this study demonstrated no positive link between muscle function and serum  $25(OH)D_3$  levels. In fact, fatigue resistance (a lower fatigue percentage) was associated with the insufficient/deficient vitamin D group. This result could be due to the increased variance in the male fatigue data, which is affected by small participant numbers, but the phenomenon remains when the male participants are excluded.

Previous studies have negatively linked muscle strength with injury incidence[23-25], with stronger dancers reporting fewer injuries[26]. This phenomenon was also observed in the current study with weaker dancers that produced less torque being significantly more likely to be injured (peak torque at 60°s <sup>-1</sup>). Muscle function profiles are similar to those previously reported in that extension peak torque and fatigue index are significantly greater than the flexion data, and peak torque at 60°s <sup>-1</sup> were greater than at 300°/s[26-28]. The lack of sex differences in peak torque at 60°s <sup>-1</sup> is unusual in our study; torque production by males is expected to be greater than females. A possible reason for this observation is the age range in the current study (13-21 years) that will encompass differing ages for adolescent pubertal development and its associated hormonal changes[29].

Injury incidence is lower than previously reported[30, 31] but is probably due to a reduced reporting period in the current study. The self-reported injury sites are similar to that previously reported with a focus on the lower limbs[31], particularly the knee and ankle. The main two types of injury were sprains and tendinopathy which amongst a number of mechanisms also have links to muscle underperformance or imbalance, through either weakness or fatigue [32, 33].

Limitations in the current study revolve around the sex imbalance in participants, the use of self-report injury history and the study's cross-sectional nature. Focused recruitment of a larger sex balanced population with equal numbers across the age range, taking into account pubertal development would provide a stronger foundation for a study. The use of prospective injury recording, either through a medical team or self-report[34] would potentially strengthen the injury data. Longitudinal data would provide an insight into vitamin D and muscle function fluctuations over the academic year and holiday periods.

## **CONCLUSION**

The current study has provided a novel insight into vitamin D levels, muscle function and self-report injuries in elite adolescent Brazilian ballet dancers. Thirty-eight percent of the cohort were classified as either insufficient or deficient despite data collection occurring during the dry season that had approximately 12 hours of sun a day. The dancers insufficient/deficient in vitamin D unusually reported greater resistance to muscular fatigue. In the current cohort, there was no link between serum 25(OH)D levels and muscular strength. A negative association between muscle strength and injury incidence highlights the need for supplemental training to rectify this risk.

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