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## ORIGINAL

### VARIATION OF BONE MINERAL DENSITY INDUCED BY EXERCISE IN POSTMENOPAUSAL WOMEN

### VARIACIÓN EN LA DENSIDAD MINERAL ÓSEA INDUCIDA POR EJERCICIO EN MUJERES POSMENOPÁUSICAS

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## ABSTRACT

**OBJECTIVE:** Assess the variation of Bone Mineral Density (BMD) in Lumbar Spine (LS) and Femoral Neck (FN). **METHOD:** 77 postmenopausal women with osteopenia. Four groups, two groups with exercise: one Estrogen/Calcitonin treatment (n=16), another supplement Calcium/Vitamin D (n=7); two groups without exercise, one Estrogen/Calcitonin treatment (n=27), and another supplement Calcium/Vitamin D (n=27). The exercise program consisted of resistance training (65% to 75% 1RM) and multi-jumps of low-to-moderate intensity. The BMD was evaluated by absorptiometry. **RESULTS.** ANOVA 2x2, homogeneity ( $p < 0,05$ ) in the BMD in both LS and FN between the pharmacological and nutritional treatments, and heterogeneity between the active and sedentary groups (LS  $p < 0,05$ , FN  $p < 0,01$ ) as also in the interaction ( $p < 0,01$ ) of the pharmacological/nutritional treatments with the physical exercise were found. **CONCLUSION.** Physical exercise program with the usual treatments, lead to a greater increase in the BMD of LS and FN in postmenopausal women.

**KEY WORDS:** postmenopause, osteopenia, mineral bone density, physical exercise, lumbar spine, femoral neck.

## RESUMEN

**OBJETIVO:** Evaluar la variación de la densidad mineral ósea (DMO) en columna lumbar (CL) y cuello femoral (CF). **MÉTODO:** 77 mujeres con osteopenia y menopausia, formaron cuatro grupos. Dos con ejercicio físico: uno *tratamiento de estrógeno/Calcitonina* (n=16), y otro *ingesta de Calcio/Vitamina D* (n=7). Dos sin ejercicio físico, uno *tratamiento estrógeno/Calcitonina* (n=27) y otro *ingesta Calcio/Vitamina D* (n=27). El ejercicio consistió en Fuerza Muscular (65% a 75% de 1RM) y multisaltos de baja a moderada intensidad. La DMO se midió mediante absorciometría. **RESULTADOS:** En ANOVA 2x2 se encontró homogeneidad ( $p < 0,05$ ) en la DMO tanto en CL y CF entre los tratamientos farmacológicos y nutricionales, heterogeneidad entre los grupos activos y sedentarios (CL  $p < 0,05$ , CF  $p < 0,01$ ) y heterogeneidad en la interacción ( $p < 0,01$ ) de ambos tratamientos con el ejercicio físico. **CONCLUSIONES:** El ejercicio físico programado junto con los tratamientos habituales conduce a un mayor incremento de la DMO de en mujeres postmenopáusicas.

**PALABRAS CLAVE:** posmenopausia, osteopenia, densidad mineral ósea, ejercicio físico, columna lumbar, cuello femoral.

## INTRODUCTION

Osteoporosis is a serious threat to the health of women of postmenopausal age, predisposing them to an increase in the risk of bone breakage. Prevention involves reducing or slowing down loss of bone tissue, maintaining bone resistance and minimizing or eliminating the factors which cause osteoporosis (1).

Hormone replacement therapy, currently considered the cornerstone of treatment for osteoporosis, is not the only alternative for adequate treatment (2). There are other forms of treatment which offer similar benefits for women at risk of osteoporosis. These include exercises in muscular resistance and dietary supplements of Calcium and Vitamin D, the aim of which is to reduce bone loss and foment new bone growth, and they have become an accepted strategy for the prevention and/or slowing down of bone loss (2,3). What is involved, therefore, is a lifestyle modification and an increase in regular physical exercise (4). Various mechanisms which reflect the beneficial effects of physical activity on bone tissue have been used for some time (5). These have a direct neurological influence, which affects vascular changes and the blood flow associated with exercise or with the mechanical and muscular tension needed to withstand exercise load (6).

The combination of pharmacotherapy and non-pharmacological treatments is vital for the success of the clinical treatment of osteopenia and osteoporosis (7). Comparable to hormone replacement therapy, physical exercise affects a multitude of systems, and therefore, may be an alternative option, since numerous studies have shown the beneficial effects of physical exercise on osteopenia via diverse physiological mechanisms (8-17).

The aim of the present study was to discover the effects of a program of muscular mass/multi-jumps physical training, with Calcium/Vitamin D treatment and Estrogen/Calcitonin, in the yearly BMD percentage variation (%) in postmenopausal women, on the lumbar spine (LS) and femoral neck (FN).

## MATERIAL AND METHOD

We studied women with osteopenia in the lumbar spine (LS) (in the L<sub>2</sub>-L<sub>4</sub> vertebral segment) or Femoral Neck (FN), who were not incapacitated for physical exercise. The sample (Table 1) comprised 77 women, forming four groups, two active groups (subjected to a program of physical exercise): one *active with intake of Estrogen/Calcitonin (AEC)* (n=16), and another *active with intake of Calcium/Vitamin D supplement* (n=7); and two non-active groups, one *non-active with estrogen/Calcitonin treatment (NAEC)*, and a *non-active group with supplementary intake of Calcium/Vitamin D (NACD)* (n=27).

**Table 1:** Mean  $\pm$  SD. Age and Years with Menopause

| GROUPS      | AGE<br>(Mean $\pm$ SD) | YEARS WITH MENOPAUSE<br>(Mean $\pm$ SD) |
|-------------|------------------------|---|
| <b>ACD</b>  | 56,57 $\pm$ 6,52       | 8,85 $\pm$ 5,27                         |
| <b>AEC</b>  | 56,12 $\pm$ 9,43       | 8,18 $\pm$ 5,87                         |
| <b>NACD</b> | 60,1 $\pm$ 10,9        | 14,96 $\pm$ 10,7                        |
| <b>NAEC</b> | 55 $\pm$ 6,45          | 8,51 $\pm$ 6,54                         |

ACD (Physical Activity and treatment with Calcium and Vitamin D), AEC (Physical Activity and treatment with Estrogen and Calcitonin), NACD (No Physical Activity and treatment with Calcium and Vitamin D), NAEC (No Physical Activity and treatment with Estrogen and Calcitonin).

Inclusion criteria: (1) Women with at least 8 years of menopause; (2) Diagnosis of osteopenia via the WHO criteria. T score between (-1) and (-2.5) (18) in the lumbar spine and femoral neck respectively; (3) Being classified as non-active women; (4) Having expressed voluntary consent in concordance with the Declaration of Helsinki; (5) Following one of the treatments, either with estrogen/Calcitonin or with Calcium/Vitamin D.

Exclusion criteria: (1) Previous fractures; (2) Functional limitations; (3) Cardiovascular illnesses or others incompatible with physical exercise.

The experiment took place over twelve months, beginning with a base bone densitometry test. The supplementary nutritional treatment and the pharmacological treatment were carried out under medical prescription: Nutritional Supplement, Calcium and Vitamin D consisted of 1000 mg Calcium/day and 400 UI of Vitamin D/day. Pharmacological, Estrogen plus Calcitonin. 0,625 mg of estrogen/day and 100 UI of Calcitonin/day during six of the twelve months which the study lasted.

The program of muscular strength training and multi-jumps was applied over twelve months, three times per week and between 60 and 75 minutes per session. It began with a 20-25 minute warm-up, which consisted of: walking, cycling or trotting at a steady pace and at low or average intensity, exercises for flexibility, muscular elasticity and joint movement. The intensity of the effort was in line with the characteristics of the sample, and varied between 2.0 to 5.9 METs, with a Reserve Oxygen Consumption (VO<sub>2</sub>R) of between 40% and 59%, equivalent to between 55 and 69% of the Maximum Heart Rate. Walking and trotting were done on a treadmill (Tredex Universal), at a speed of between 4.0 and 8.5 km/h. The exercise on the cycle ergometer (Monark), was done with a fixed mechanical resistance of between 0.5 and 1.0 Kiloponds (Kp) at a speed of between 50 and 60 rpm.

In the main part of the training we used single joint and multi-joint exercises for muscle strength at an intensity which varied between 65 and 75% of 1MR (Maximum Repetitions) of Maximum Dynamic Force (MDF). The MDF was calculated via a 8-10MR sub-maximum evaluation test. The development of muscle strength was carried out with exercises for concentric-eccentric muscle contraction; 3-5 series, 8 to 10 repetitions for each muscle group, with one minute cool downs.

The physical work of multi-jumps was low to moderate intensity, with vertical, horizontal and platform jumps with immediate response from a low height (30cm) and onto a semi-soft surface, without overload, alternating between 1 or 2 legs and with knees bent at a falling angle of between 130° and 150°.

| Exercise Program: Multimodal Physical Training                               |   |  |   |
|--|---|--|---|
|  | Day 1   | Day 2  | Day 3   |
| <b>WARM UP</b>   | 1. Walking (5 min). 4-6 Km/h<br>2. Flexibility (5 min)<br>(major muscles and joints)<br>3. Smooth running (10 min)<br>(uniform rhythm/speed changes<br>7,0 a 8,5 km/h)                                    | 1. Stationary bicycle. (10 min), 50-60 rpm<br>2. Flexibility (5 min)<br>3. Smooth running (10 min)<br>(uniform rhythm/speed changes 7,0 a 8,5 km/h))                           | 1. Walking (5 min) 4-6 Km/h<br>2. Flexibility (5 min)<br>(major muscles and joints)<br>3. Smooth running (10 min)<br>(uniform rhythm/speed changes 7,0 a 8,5 km/h)  |
| <b>PRINCIPAL</b>   | 1. Leg press*<br>2. Curl de biceps*<br>3. Multi-jumps<br>(1-3 series, 10-15 rep)<br>4. Dumbbells bench press *<br>5. Leg extension*<br>6. Multi-jumps<br>(1-3 series, 10-15 rep)<br>7. Pulley pull downs* | 1. Leg extension*<br>2. Curl de biceps*<br>3. Multi-jumps<br>(1-3 series, 10-20 rep)<br>4. Dumbbells bench press*<br>5. Leg press *<br>6. Pulley pull downs *<br>7. Leg curl * | 1. Leg curl*<br>2. Lateral dumbbell flies*<br>3. Multi-jumps<br>(1-3 series, 10-20 rep)<br>4. Curl de biceps*<br>5. Leg extension*<br>6. Pulley pull downs *<br>7. Multi-jumps<br>(1-3 series, 10-20 rep) |
| <b>COOL DOWN</b>   | Flexibility   | Flexibility  | Flexibility   |
| Time: 60 -75 minutes/session. Program Length : 12 months<br>*65 al 75% (1RM) |   |  |   |

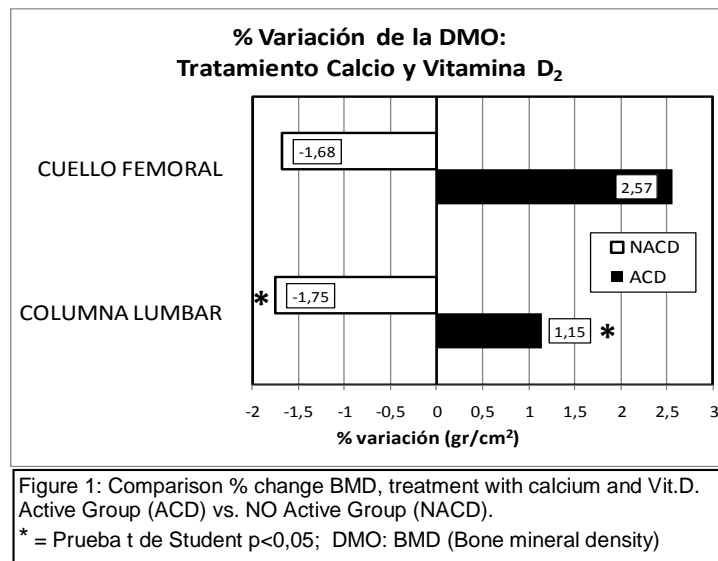
The degree of osteoporosis was determined by bone densitometry, via a dual source of X rays, which show the physical properties of the bone and soft tissue. We used a Norland XR-26 dual energy absorptiometer with dual X ray detectors, with a Samario filter system for level K. The results obtained were calculated using an XR-26 IBM PS-2 computer, which registered the average bone density of the

lumbar spine L<sub>2</sub>-L<sub>4</sub> and the bone density of the femoral neck (FN) of the left hip in grams per square centimetre (gcm<sup>2</sup>).

For the inferential statistical analysis we used the ANOVA Factorial 2x2 dual variance analysis to analyse the effects caused by different levels of the two factors and the Student t test to compare the results.

## RESULTS

Figure 1 represents the percentage of annual variation in BMD, in the two groups that were treated with Calcium and Vitamin D, Group ACD and Group NACD.



In the active group with Calcium and Vitamin D supplement (ACD), we observed a significant increase in the annual variation of BMD, both in FN and LS, with LS statistically significant ( $p < 0,05$ ). We also observed a notable decrease in the annual variation of BMD in the non-active group with Calcium and Vitamin D supplement in both bone structures, with the lumbar spine statistically significant.

Figure 2 shows the comparison percentage change in annual variation of BMD between the AEC Group and the NAEC Group. Both groups were treated with Estrogen and Calcitonin. No statistically significant results were found in either of the groups in either of the bone areas analysed.

Table 2 shows the results of the application of the ANOVA 2x2 on the annual percentage variation of BMD in Lumbar Spine. In the *BETWEEN ROWS* contrast, we found no statistically significant differences at a 95% confidence level, in the average percentages of the annual BMD variation in LS (L<sub>2</sub>-L<sub>4</sub>), between the

women treated with *estrogen/calcitonin versus Calcium/Vitamin D*, with homogeneity in the variation of BMD between the different treatments, both nutritional and pharmacological in LS.

**Table 2.** 2x2 FACTORIAL ANOVA B.M.D. in Lumbar Spine.

| Variation Source                                       | SS     | df | MS    | F         |
|--|--------|----|-------|-----------|
| <b>Between Rows (Ca-VitD/ Estrog-Calcitonin) (VII)</b> | 10,38  | 1  | 0,38  | 1,73 (NS) |
| <b>Between Columns (Active/NO active) (VI2)</b>        | 35,68  | 1  | 35,68 | 5,94*     |
| <b>Interaction (VII*VI2)</b>                           | 48,96  | 1  | 48,96 | 8,16**    |
| <b>Error</b>   | 311,90 | 52 | 6,0   |           |
| <b>Total</b>   | 407,00 | 55 |       |           |

\* (p<0,05); \*\* (p<0,01); (NS) Not significant

In the *BETWEEN COLUMNS* comparison, which contrasts the annual variation percentages of BMD in LS (L<sub>2</sub>-L<sub>4</sub>) between the groups with physical activity versus the groups without physical activity, we observe that independently of treatment with estrogen/Calcitonin or Calcium/Vitamin D, the different statistic found in the groups was significant (p<0.05) when the program of physical training was taken into consideration.

Table 3 shows the results of the ANOVA 2x2 comparison of the annual variation percentage of BMD in FN. In the *BETWEEN ROWS* contrast for FN we found no significant differences when we compared the treatments with *estrogen/Calcitonin versus Calcium and Vitamin D supplement*. In the *BETWEEN COLUMNS* comparison of the annual variation percentage of BMD in LS in *active women versus non-active women*, independently of nutritional or pharmacological treatment, the results were statistically significant (p<0.01), which showed that the presence of physical activity produced positive increases in BMD of the FN in the women studied.

**Table 3.** 2x2 FACTORIAL ANOVA B.M.D. in Femoral Neck

| Variation Source                                       | SS      | df | MS      | F         |
|--|---------|----|---------|-----------|
| <b>Between Rows (Ca-VitD/ Estróg-Calcitonin) (VII)</b> | 19,32   | 1  | 19,32   | 1,75 (NS) |
| <b>Between Columns (Active/NO active) (VI2)</b>        | 77,55   | 1  | 77,55   | 7,02**    |
| <b>Interaction (VII*VI2)</b>                           | 121,478 | 1  | 121,478 | 11,00**   |
| <b>Error</b>   | 573,984 | 52 | 11,04   |           |
| <b>Total</b>   | 792,332 | 55 |         |           |

\* (p<0,05); \*\* (p<0,01); (NS) Not significant

## DISCUSSION

Different studies state that Calcium supplements do not always significantly reduce the risk of fracture in postmenopausal women, such as, for example, femoral neck fracture. Five large scale studies, randomized and controlled, have raised questions as to the benefits of Calcium for the reduction of risk of fracture, (19). These results are in concordance with the results of this paper regarding the scores of BMD in the Femoral Neck, where the groups who underwent this treatment for 12 months show no significant changes (Figure 1). We should add here that the number of subjects of the ACD group who completed the research was small, which could influence the statistical strength of these results. However, there is a marked tendency to the opposite in the Active Group (ACD), whose scores increased in relation to the Non-Active Group (NACD) in which the BMD decreased.

This different tendency between the ACD and the NACD groups is also reflected in the results for BMD in FN (Figure 1), but in this case, the increases in BMD in the active group reflect statistically significant differences.

Some studies have examined the effect of different doses of Calcium and Vitamin D, alone or combined, to increase bone mineral density (20) and it has been shown that Calcium plus Vitamin D reduces or detains bone loss in healthy postmenopausal women and in postmenopausal subjects with substantial bone loss or with previous fracture. As can be seen in Figure 1, the levels of BMD in the NACD group hardly changed at all in our study. We should state that the average age is higher than that of the active group, so this factor could influence these results. In this sense, in a study of 36,282 women inscribed in the Women's Health Initiative (WHI), who received a daily dose of 1000mg of Calcium Carbonate with 400 IU/day of Vitamin D, during an average monitoring period of 7 years, the age range was from 50 to 79 years. Small improvements in bone mineral density of the Femoral Neck (FN) were also found, yet while they were significant in this case, proportional reduction of the risk of hip fracture was not found (21).

Estrogen plays an important role in regulating the balance between bone formation and reabsorption, as well as the stimulation of osteoblast activity (22). It has shown greater beneficial effects on BMD of the bone when used in conjunction with strength training exercises, than when hormonal therapy alone is used (8, 17). Results were found in this study which are in concordance with the findings of the studies cited, although while the results are not statistically significant, there is an improvement in the BMD of the Lumbar Spine in the two groups which underwent estrogen and calcitonin hormone treatment (Figure 2).

In the ANOVA 2X2 BETWEEN COLUMNS comparison of the annual variation (%) of BMD in LS between the Active Groups and Non-Active Groups, and taking into



consideration the training program as a differentiating factor, we see that there are statistically significant differences ( $p < 0.05$ ), independently of the treatment with estrogen/Calcitonin or Calcium/Vitamin D (Table 2). These results vary from other studies which have highlighted the little effect of exercise programs on bone mass in pre- and postmenopausal women (12). However, the findings coincide with the results of other meta-analyses which confirm that physical exercise has a positive effect on the increase on bone mineral density (23).

Through an analysis of the results of *INTERACTION*, we observe a significant percentage variation ( $p < 0,01$ ) in bone mineral density in FN (Table 2) when treatments with Estrogen/Calcitonin and Calcium/Vitamin D supplement with levels of *physical activity* are related. These differences indicate that treatments with Estrogen/Calcitonin and Calcium/Vitamin D supplement are more effective when combined with physical activity, producing significant changes in the increase of trabecular BMD, showing clear that the program of applied training appears to have a synergistic effect with nutritional and pharmacological supplement treatment for the increase in BMD in LS in postmenopausal women

In the ANOVA 2X2 *BETWEEN ROWS* contrast for FN, significant differences were not found in the variation of BMD on FN when comparing treatments with *Estrogen/Calcitonin* versus the *Calcium and Vitamin D* supplement (Table 3). Evidence in the reduction of risk of fracture is less clear for the Calcium and Vitamin D treatment (24, 25). Some meta-analyses reported a reduction in risk for standard Vitamin D in comparison with the placebo (26-29), while others do not coincide in their conclusions (30, 31). In the same way, WHI showed that estrogen significantly reduced the risk of hip fracture (32). However, in a meta-analysis which included data from the WHI there was a non-significant reduction in risk of hip fracture (31). The results from this study coincide with Campusano et al. (33) in that there is no guarantee that the use of Calcitonin in doses of 100 UI 3 times/week or less, as frequently occurs in clinical practice, will increase bone remodelling in postmenopausal women

In the *BETWEEN COLUMNS* contrasts, which compared annual variation (%) of BMD in FN of the *Active Groups* versus *Non-Active Groups* (Table 3), independently of nutritional or pharmacological treatment, the results were statistically significant ( $p < 0,01$ ), highlighting heterogeneity in bone variation and showing that the presence of physical activity in the training program, by itself, caused positive increases in BMD of the FN in the women studied. In the light of the results we found, this type of exercise seems to be an important factor in the prevention of osteoporosis and the reduction of risk of fracture, since in addition to improving muscular strength, balance and general mood are improved (12).

In this study we have seen that improvement in BMD of the FN was aided by the physical exercise program and its effect on the conservation or increase of muscle

mass (13). Specificity of load, the means and methodology of the strength training based on moderate to high intensity physical stimulus with little repetitiveness, along with multi-jumps, subjected the body to a mechanical stress greater than that inherent in daily life, with the aim of fomenting greater bone mineral density (34). These results contrast with those of low impact activities, such as walking, swimming, etc. (35, 36). It can be inferred that these physical stimuli applied in a specific and controlled way acted on the bone tissue, which is sensitive to stress and to moderate to high intensity impact, contributing to the transitory deformation of the bone necessary to supply a sufficient amount of mechanical stress to stimulate the growth of the new bone (16). Findings similar to those of this study confirm that the later effects of a long-term multicomponent exercise program, of eleven months duration, led to an increase in T-score of the femoral neck in postmenopausal women with low bone density increasing notably in the exercise group, and decreasing significantly in the control group, who did no exercise (11).

Finally, the result of *INTERACTION* in the variation (%) of annual bone mineral density in FN, compared with the *estrogen/Calcitonin treatment, Calcium/Vitamin D supplement with the level of physical activity* (Table 3), shows that the significant differences ( $p < 0,01$ ) observed are induced by the physical activity variable, which influenced the changes in the BMD of the FN much more than each action of each nutritional or pharmacological treatment by itself. The results therefore show an increase in the annual variation of BMD of the FN in active women with Calcium/Vitamin D. These results do not concur with some studies which examine the effectiveness of muscle resistance exercise on bone mineral density (BMD) in femoral neck (FN) in premenopausal women, the results of which do not support the effectiveness of muscle resistance exercise in increasing or maintaining mineral density in FN in premenopausal women (14). However, we find a generalized lack of description of the methodology employed, for example, concerning the basic description parameters of the stress load applied. This lack, in these studies, means that we cannot compare the real benefits of physical exercise.

Studies deal with the effects of stress load of different physical protocols on bone formation, affirming that dynamic, rather than static load, is more effective in bone formation (16); that the bone formation rate is positive when related to loading frequency (37). Also, the frequency of exercise sessions and the inclusion of rest periods significantly increases bone formation in comparison with continuous loading cycles (38), while also that bone tissue becomes quickly insensitive to prolonged exercise (39). But most of these studies fail to detail the program of physical exercise applied, nor their qualitative or quantitative characteristics. Training loads must be localized and specific for the target osteo-muscular structure to favor osteogenesis, due to the specificity principles of stimulation (40). Physical stimulation which must be chronic and exceed the usual overload in order to remodel bone tissue (41).

## CONCLUSIONS

The results found in this study support the adjunctive role of multi component exercise with a high percentage of muscle strength/multi-jumps in order to increase BMD of the FN and significantly in LS, independently of the treatment applied.

In accordance with the results in this paper, it can be stated that the effects of exercise on bone density were more homogenous in body regions with a greater trabecular bone, found especially in the vertebral bodies of the lumbar vertebrae.

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