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Are serum calcium and vitamin D levels effective in osteoporosis?

Serum kalsiyum ve D vitamini düzeyleri osteoporozda etkili midir?

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ABSTRACT

The reason by we work to compare serum calcium and vitamin D levels of osteoporotic and non-osteoporotic patients.The study included a total of 181 non-osteoporotic individuals and 94 osteoporotic patients over 50 years of age, post-menopausal, who referred to the gynecological clinic at our hospital and diagnosed with osteoporosis following a Dual Energy X-Ray Absorptiometry (DEXA) after their serum calcium and vitamin D levels were checked by laboratory analyses. Both groups were comprised of patients who did not receive any medication due to a chronic disease or osteoporosis. Descriptive statistics and graphs, as well as statistical analysis (Chi-Square Test, Independent Samples T-Test, One-Way ANOVA Test and Pearson's Correlation) were used in the study. These analyses were calculated at a 95% confidence level. SPSS 19.0 (Statistical Package for the Social Science) was used for analysis and calculation of descriptive statistics. Both groups were compared individually in terms of age, body mass index (BMI), gravida, parity, abortion, curettage, age of menarche, age of menopause, duration of menopause, smoking and alcohol consumption, total lumbar T score, femoral neck T score, serum calcium and vitamin D. Demographic data and clinical features were statistically similar in both groups; however, total lumbar T score and femoral neck T score were significantly lower in the osteoporotic group. There was no significant association between age and osteoporosis. (χ^2=3.381, p=0.184). There was no significant difference in vitamin D levels between age groups (F=0.552, p=0.576). The peer groups presented no significant difference in terms of calcium levels (F=0.716, p=0.490). There was no significant difference in vitamin D levels between osteoporotic and non-osteoporotic groups as well (t=-0.83, p=0.934). The groups showed no significant difference in terms of calcium levels (t=0.587, p=0.564). No association was detected between the measured vitamin D and calcium levels (Pearson's Correlation: 0.049, p=0.416).To conclude , we can use serum calcium and vitamin D levels together with other methods that provide information about whether patients have osteoporosis. In light of this information, we can begin prophylaxis or early treatment of osteoporosis, which is currently a common disease that leads to morbidity and mortality.

Keywords: Osteoporosis, menopause, calcium, vitamin D.

ÖZET

Bu çalışmadaki amacımız Osteoporoz tanısı alan ve almayan hastaların serum kalsiyum ve D vitamini düzeylerini karşılaştırmaktır. Hastanemiz jinekoloji polikliniklerine başvuran postmenopozal, 50 yaş üzeri, serum kalsiyum ve D vitamini düzeylerine bakılmış, Dual Enerji X-Ray Absorbsiyometri (DEXA) uygulanması sonucunda 94 adet osteoporoz tanısı almış, 181 adet osteoporoz tanısı almamış ve osteoporoz nedeniyle herhangi bir ilaç tedavisi almayan hastalar çalışmaya alındı. Çalışmada tanımlayıcı istatistikler ve grafikler, istatistiksel analizler (Chi-Square Test, Independent Samples T-Test, One-Way Anova Test ve Pearson Correlation) kullanılmıştır. Bu analizler %95 güven seviyesinde hesaplanmıştır. Analiz ve tanımlayıcı istatistiklerin hesaplanmasında SPSS 19.0 (Statistical Package for the Social Science) kullanılmıştır. Eldeki verilerle yaş, kemik durumu, serum kalsiyum ve D vitamini kendi aralarında tek tek karşılaştırıldı. Demografik veriler ve klinik özellikler her iki grupta istatiksel olarak benzerdi. Yaş ile Osteoporoz durumu arasında istatistiksel açıdan anlamlı bir ilişki bulunmadı. (χ^2=3.381, p=0.184). Yaş grupları arasında D vitamini açısından istatistiksel açıdan anlamlı bir farklılık gösterilemedi(F=0.552, p=0.576). Yaş grupları arasında kalsiyum açısından istatistiksel açıdan anlamlı bir farklılık bulunmamaktadır(F=0.716, p=0.490). Osteoporoz olan ve olmayan gruplar arasında D vitamini değeri açısından istatistiksel açıdan anlamlı bir farklılık görülmedi. (t=-0.83, p=0.934) Heriki grup arasında kalsiyum değeri açısından istatistiksel açıdan anlamlı bir farklılık yoktu. (t=0.587, p=0.564).Ölçülen D vitamini değeri ile Kalsiyum değeri arasında ilişki tespit edilemedi (Pearson Correlation:0.049, p=0.416).Sonuç olarak Serum kalsiyum ve D vitamini düzeyleri, hastaların osteoporoz olup olmadığı hakkında bilgi veren diğer yöntemlerle beraber kullanılabiliriz. Bu yolla edinilen bilgiler ışığında günümüzde sık karşılaşılan,morbidite ve mortaliteye yol açan osteoporoz hakkında profilaksi veya erken tedaviye başlayabiliriz.

Anahtar Kelimeler: Osteoporoz,meonopoz, kalsiyum, vitamin D.

1. INTRODUCTION

Osteoporosis (OP) is a structural failure of bone, characterized by low bone mass and degradation of the micro-architecture of bone tissue. Increased bone fragility in the OP also leads to an increase in morbidity and mortality (Rodríguez and García, 2002). The World Health Organization (WHO) has defined OP based on Dual Energy X-Ray Absorptiometry (DEXA) measurements. With DEXA, which was introduced in 1987, Bone Mineral Density (BMD) measurement has been recognized as the gold standard for the identification and evaluation of osteoporosis and is the most widely employed technique in clinical practice. Therefore, it also plays an important role in the prevention of bone fractures, which is the most important result of OP. In order to diagnose OP and determine the risk of fracture, OP risk factors need to be investigated and bone mineral density should be evaluated with DEXA in their presence (Sosa and Gómez, 2009). Osteoporosis risk factors include white race, female sex, low body mass index, occupation, menopause, history of previous fragility fracture, consumption habits such as tea, coffee, smoking, sedentary lifestyle, chronic diseases, medical conditions such as steroid use, cognitive functions and personal characteristics such as mood disorders that cause a tendency to fall (Lips P, 2001). Vitamin D has a central role in stimulating calcium absorption. Bone loss in women is accelerated by the loss of estrogen in menopause. Although the effects of vitamin D on bone mineral loss during menopause are still controversial, it has been shown that vitamin D intake during menopause reduces this loss. For this reason, vitamin D and calcium levels may inform us about whether patients develop osteoporosis (Reginster, 2005).

It is important to identify OP risk factors in terms of preventive treatment and to prevent OP-related morbidity and mortality with early diagnosis by identifying particularly at-risk patients (Chen et al., 2007). Although most studies have investigated the relationship of risk factors with OP and associated fracture incidence rate, there are a limited number of studies that investigate whether calcium and vitamin D are associated in osteoporotic patients diagnosed by DEXA measurements and non-osteoporotic individuals. Therefore, in this study, we aimed to evaluate whether Calcium and Vitamin D levels were associated between osteoporotic patients diagnosed by DEXA level and non-osteoporotic individuals.

2. MATERIAL AND METHOD

The study included a total of 275 post-menopausal women who referred to our gynecological outpatient clinic from January 2016 to December 2018 and diagnosed with OP

for the first time according to the WHO criteria. None of these patients had received any treatment for osteoporosis, including herbal calcium supplements, calcium preparations, vitamin D and alendronate, which was the main criteria for inclusion in the study. Patients whose total lumbar or femoral neck T score was below -2.5, as assessed by DEXA, comprised the study group. Other post-menopausal patients with no osteoporosis were included in the control group. The study was approved by the Local Ethics Committee. Universal principles of the Helsinki Declaration were applied to the study. Patients with significant gynecological or medical complications and those with unreliable information were not included in the study. Patients with a known organic brain injury such as dementia, Alzheimer's disease, Parkinson's disease, those with a history of ischaemic or haemorrhagic cerebrovascular disease, medicated or non-medicated patients with a diagnosed psychiatric disorder, those patients whose DEXA measurements were performed at any medical center out of our hospital, those who received anticoagulants such as heparin and HRT treatment were excluded from the study. Patients with metabolic, endocrine, connective tissue diseases, history of fragile fractures, chronic disorders such as chronic liver disease, malabsorption syndrome, and those who received medication that may cause secondary OP such as corticosteroids, anticonvulsants, thyroid hormone, diuretic and methotrexate were also excluded from the study. All of the patients' BMD and biochemical markers were evaluated in detail by the same person. Every patient was included only once, following referrals were not included in the study. Interviews with patients as well as computer-based patient records reviewed in a comprehensive manner were recorded for the following information: patients' age, height, weight, gravida, parity, abortion and/or curettage, the first menarche age, age at menopause, and duration, educational background, income level, smoking and alcohol consumption, total lumbar and femoral BMD and levels.

2.1. BMD Assessment

BMD measurements were performed at the radiology department of Tepecik Training and Research Hospital, University of Health Sciences. As part of the study, the lumbar spine (L1-L4 and L2-L4) and proximal femur (femur total, femur trochanter and Ward’s triangle) BMDs of the patients were measured anteroposterior using DEXA method and Hologic device (Hologic, ASY-05119, 2014-Source, Inc: Bedford, USA). The scan voltage was 80 kv, 1500 mA current, at a dose of 21.0 μGy. The scan time was about 3 minutes. The results were evaluated by taking BMD (g/cm2), T and Z scores of both regions. T score = Measured BMD-young adult average BMD/ young adult normal SD (SD: standard deviation) Z score =

Measured BMD-same age group average BMD / same age group SD. Although the Z score is not as clinically valuable as the T score, deviations from the normal in the Z score necessarily require a detailed investigation of the patient for metabolic bone diseases and secondary osteoporosis causes. Various epidemiological studies have shown that when the bone mineral density at any anatomic region is one standard deviation (SD) less than the average bone mineral density of the young adults in the society, this at least doubles the risk of future hip fracture (Simonelli et al., 2008). Since the WHO's publication in 1994, most clinicians and researchers have identified osteoporosis as a bone mineral density of 2.5 SD or lower than the young reference society average bone mineral density, based solely on bone mass measurement. This acknowledged threshold is useful for prevalence studies and for identifying cases with a high risk of fracture. In our study, we also evaluated DEXA measurements according to the T score (Boonen et al., 2005). We took a T score >-1 as normal and low fracture risk, T score between -1> and >-2.5 as above-average fracture risk and osteopenia, <-2.5 as osteoporosis and high fracture risk, and T score <-2.5 and presence of fracture as very high risk of fracture. Body mass index (BMI) = weight (kg)/height (m2) was calculated by measuring women's height and weight.

2.1.1 Laboratory analysis

Blood samples were taken at our hospital lab from the volunteers to gel and clot activator tubes (BD Vacutainer SST II Advance, 5 mL, 13x100 mm, catalog number 367955, Becton Dickinson, NJ, USA) after at least 8 hours of fasting. After waiting for 30 min, the blood samples were then centrifuged at room temperature for 10 minutes to obtain serum. Hemolytic, icteric, or lipemic samples were excluded from the study. The samples were measured for calcium (Ca2+) by the spectrophotometric method in the Beckman Coulter AU-5800 autoanalyzer in the Central Biochemistry Laboratory of Tepecik Training and Research Hospital. The 25 (OH) D measurements were performed using the chemiluminescence immunoassay method in the Advia Centaur XP analyzer (Siemens Healthineers, Erlangen, Germany).

2.1.2 Statistical analysis

The data were analyzed using the Statistical Package Social Sciences (SPSS), version 19.0 (SPSS Inc., Chicago, IL, USA). Descriptive statistics were shown as mean±standard deviation for continuous variables and as observation number and (%) for nominal variables. Descriptive statistics and graphs, as well as statistical analysis (Chi-Square Test, Independent Samples T-Test, One-Way ANOVA Test and Pearson's Correlation) were used in this study.

These analyses were calculated at a 95% confidence level. P<0.05 was taken to indicate statistical significance.

3. RESULTS

The study was conducted including a total of 275 women with and without osteoporosis. The analyses were performed for 2 groups with and without osteoporosis. Osteoporotic women accounted for 34.2% (n=94) of the population, while non-osteoporotic women comprised 65.8% (N=181). Patients' demographic and clinical characteristics are shown in Table 1. The two groups presented no significant difference in terms of age, BMI, gravida, parity, abortion and/or curettage, the first menarche age, age at menopause and duration, smoking and alcohol consumption (p: 0.02, 0.03 and 0.04, respectively). Total lumbar T score and femoral neck T score were significantly lower in the osteoporotic group. Subjects' educational background, income levels and smoking habits are compared in Table 2. There was no statistically significant difference between study and control groups in terms of educational background, income levels or smoking habits (p: 0.23, 0.32 and 0.24, respectively). The average age (51-88) of the women who participated in the study and whose osteoporosis levels were calculated was 65.07 ± 8.68. Table 3 shows the age distribution of women. Of the women with osteoporosis, 28.7% (n=27) were 60 years of age and under, 41.5% (n=39) were 61-70 years of age, and 29.8% (n=28) were older than 70. Of non-osteoporotic women, 39.8% (n=71) were 60 years of age and under, 36.5% (n=66) were 61-70 years of age, and 23.8% (n=43) were older than 70.

There was no significant relationship between age and osteoporosis ($\chi 2$=3.381, p=0.184). Also, there was no significant difference in vitamin D levels between peer groups (F=0.552, p=0.576). The peer groups indicated no significant difference in terms of Calcium levels (F=0.716, p=0.490). There was no significant difference in vitamin D levels between study and control groups as well (t=-0.83, p=0.934). The groups presented no significant difference in terms of Calcium levels (t=0.587, p=0.564). No association was detected between the measured vitamin D and calcium levels (Pearson's Correlation: 0.049, p=0.416).

4. DISCUSSION

Osteoporosis is a common, chronic, progressive and systemic disease characterized by decreased bone density and increased bone fragility due to deterioration in the microstructure of bone tissue, affecting millions of postmenopausal women as well as men at various ages (Fordham, 2004). In Europe and North America, osteoporosis has been detected in 6% of men

aged 50-84 and 21% of women. Factors such as low peak bone mass size reached by women relative to men and low mineral content, as well as increased bone loss associated with the deficiency of estrogen lead to OP differences between the two sexes. Also, being female is reported as a major risk factor (Schmitt et al., 2009). Therefore, we evaluated only post-menopausal women of similar age to ensure homogeneity in our study.

According to the WHO-recommended and widely used definition, menopause is the permanent termination of menstruation as a result of the “loss of ovary activity." In this period, following the risks associated with the cardiovascular system, the highest risk for women is the loss of bone tissue and associated osteoporosis. In women, the cessation of ovarian function during menopause and the cessation of estrogen production accelerates bone loss in connection with age and increases the severity of osteoporosis. The goal of preventing osteoporosis is to maximize bone mass. Bone mass reaches its maximum density at 30-35 years of age (Schmitt et al., 2009). After this age, the bone cycle results in loss of bone mass. The higher the bone mass, the lower the risk of developing osteoporosis in advanced age. In this respect, it is necessary to take measures to increase bone mass from childhood. Age, sex, genetic factors, nutrition, hormonal changes, alcohol consumption, exercise, the first menstrual age and age at menopause are factors that affect osteoporosis (Gallagher, 2007). Female and male sex hormones prevent bone loss by inhibiting the activity of osteoclasts and stimulating new bone construction of osteoblasts. Given that age, sex and genetic factors are not modifiable, nutrition and exercise appear to be important factors in preventing osteoporosis. While calcium and vitamin D intake and regular exercise reduce osteoporosis, smoking, alcohol, tea and coffee consumption has an increasing effect on osteoporosis (Lombardi et al., 2004).

Osteoporosis is asymptomatic in many individuals until fracture development; early diagnosis is important in this respect. Bone mineral density is measured in the diagnosis and follow-up of osteoporosis (Boonen et al., 2005). For this purpose, the anteroposterior DEXA method as is used as the gold standard. Osteoporotic fractures cause significant medical and financial losses and severely impair the quality of life. In this respect, it is important to recognize osteoporosis in order to raise women's awareness of osteoporosis and to carry out studies for preventive measures. A patient who is suspected of having osteoporosis should definitely be evaluated in terms of these risk factors, ten-year fracture risk should be determined, and DEXA measurements and treatment should be planned according to these findings (Boonen et al., 2005). Although osteoporosis is defined by WHO according to

DEXA measurements, it is reported in the literature that individual characteristics that increase the likelihood of OP development in patients and help identify those prone to OP, i.e. existing OP risk factors, should also be evaluated (Lips at al., 1987). Early detection of risk factors will help patients to receive early diagnosis and allow for the planning of treatment. However, studies that investigated the relationship of these risk factors with OP reported conflicting results. There are currently a limited number of studies evaluating the relationship between risk factors and DEXA levels. However, since these risk factors include a very broad range of characteristics, we considered only the effects of calcium and vitamin D by equalizing these risk factors to the maximum extent, rather than investigating all known risk factors.

Vitamin D in a normal level is essential not only for optimal bone development but also for the prevention of many chronic diseases. Vitamin D deficiency is a defined risk factor for osteoporosis, falls and fractures. Along with deteriorated bone formation, vitamin D deficiency causes proximal myasthenia and impaired neuromuscular coordination, increasing the risk of fractures and susceptibility to falls. The negative effects of fracture-associated pain and functional restriction on quality of life are well known (Holick et al., 2011).

It has been shown in many studies that malnutrition, decreased vitamin D synthesis in the skin and decreased utilization of sunlight are the most important causes of vitamin D deficiency in the elderly and this is an important risk factor for hip fracture which is the major cause of morbidity and mortality in old age (Lips and van Schoor, 2005). Less than half the patients ' mobility can be restored one year after the fracture and approximately 20% are lost (Sernbo and Johnell, 1993). As long as the serum vitamin D level is adequate, the body can adapt even to very low calcium intake. Calcium absorption decreases with menopause and advancing age. This may be related to increased intestinal vitamin D receptor resistance to 1.25 (OH)2 vitamin D and decreased synthesis of 1.25 (OH)2 vitamin D (Ledger et al., 1994). Studies with vitamin D supplements found a positive correlation between serum calcidiol levels and lumbar vertebral BMDs in post-menopausal women and femoral neck, thoracanter and Ward’s triangle BMDs in women over 60 years of age (Martinez et al., 1994). Studies in older women on vitamin D and calcium supplementation have shown that vitamin D supplementation causes increased BMD in the femoral head with relatively high cortical bone and reduces the rate of non-hip and non-vertebral fractures (Holick et al., 2011). Given the literature on osteoporosis, it seems logical, as an idea, that serum calcium and vitamin D decrease with advancing age may be associated with osteoporosis, which is an important

public health problem. It is emphasized that regular calcium intake may be beneficial in preventing osteoporosis. In a study conducted with 177 osteoporotic women over the age of 65, it was reported that calcium supplement therapy alone is not sufficient to improve the quality of life, response to treatment is very limited after the development of osteoporosis, bone loss must be stopped by determining at-risk patients at an early stage and that adequate consumption of calcium by diet is required (Gulseren et al., 2014). Some studies have demonstrated that there is a positive relationship between dietary calcium and bone mineral density, while some researchers have reported that there is no such relationship (Chen et al., 2007). Pinar et al. detected an inverse relationship between daily protein intake and osteoporosis but found no statistically significant difference (Pınar et al., 2009). Lukert et al. have shown that vitamin D intake during menopause reduces bone mineral loss (Lukert et al., 1992). In our study, we did not find a significant relationship between vitamin D and calcium intake and osteoporosis.

Low vitamin D levels, decreased calcium absorption, increased bone resorption with increased levels of PTH and bone loss are involved in the development of osteoporosis. In our study, there was no significant difference between patients lacking vitamin 25(OH) D and calcium levels and osteoporosis. In clinical practice, Holick et al. recommended that only vitamin D levels be looked at in risk group patients. These indications include rickets, osteomalacia, osteoporosis, chronic kidney failure, liver failure, Crohn's disease, hyperparathyroidism, medication (glucocorticoid, antiepileptic, antifungal, etc.), pregnant and breastfeeding women, elderly patients with a history of falls and non-traumatic fractures, obese children, individuals with body mass index >30kg/m², sarcoidosis and some patients with lymphoma (Chapuy et al., 1992). Since the measured calcium and vitamin D values reflect acute values, they are not considered sufficient to make a precise evaluation in retro projection. Additional studies are needed to prove that osteoporosis can be used as additional guidance to the diagnostic criteria. Although the factors that may be associated with osteoporosis have been minimized in our study, further studies are needed to explain the similar occurrence of calcium and Vitamin D levels in the study and control group, with the same standards or more detailed and significant reduction of risk factors.

In our study, patients with similar Body Mass Index (BMI) were included for the homogenization of the groups. Therefore, no significant difference was found between the groups in terms of BMI. Low BMI is a risk factor for hip and all osteoporotic fractures, but it is a protective factor for lower leg fractures. High BMI, on the other hand, is a risk factor for

upper arm (humerus and elbow) fractures (Wardlaw, 1996). It is important to have a BMI within the recommended limits to protect against osteoporosis.

The present study offers a couple of further contributions to the literature. First, it is possible to say that we performed the comparison in a homogeneous picture despite a retrospective design: While selecting the cases, it was designed in such a way to not affect the study's findings based on a prior meticulous examination procedure regarding patients' age and BMIs. All subjects were analyzed by the same DEXA device and with the same biochemical instruments during the study period and for the whole study group. Moreover, another factor that increased the quality of our study was our attempt to equalize many gynecological and medical conditions with the potential to influence the parameters studied. All this caused our study population to be numerically inadequate. A larger population could yield statistically significant results as it would increase the consistency of the data. It is possible to have statistically significant results if many other parameters such as collecting information about the bone metabolism in the family, the use of drugs affecting metabolism, the use of drugs with the potential to change the bone density, the condition of exercise and the type of exercise, if the patient performs any exercise. Moreover, it is less likely for this factor to affect the study findings because patients with unreliable files and information were excluded from the study. The present study also has certain limitations that should be noted: It was conducted at a single institution. Another inference from this study is that calcium and vitamin D levels vary in lifestyle and from region to region. The area where the study was conducted is in a sunny location. Since this situation was observed to affect both the study and control group endocrinologically during the study period, similar studies should be carried out in the regions that have a broad range of climates. The performance of further multi-centered studies on this issue may consolidate our findings.

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