

Investigating the levels of osteocalcin in the blood serum and measuring its levels in patients with osteoporosis in Al-Qadisiya Governorate-Iraq

Ahmed Issa Abdul Hussain¹

Dr. Ali Abdul Hussein Ghazaly^{*1}

¹ Department of Biology, College of Science, University of Al-Qadisiya, Iraq. *Corresponding Author

Abstract

Osteoporosis is a major public health problem because it leads to weakness of skeleton, increase risk of fractures particularly of the spine and hip, increases morbidity and mortality which are a huge burden on the health system. Each bone consists of a thick outer shell known as cortical bone and a strong inner network of honeycomb-like trabecular bone, with blood and bone marrow between the bone supports, so osteoporosis occurs when the supports that make up this bone. The skeleton becomes thin, causing the bones to become brittle and break easily. Osteoporosis is usually associated with postmenopausal women, but it can affect younger men and women, children and pregnant women as well. Known causes of low bone density include estrin deficiency, nutritional deficiency (low intake of calcium and vitamins D and K), limited physical activity, "weak" syndrome, and nicotine or excessive alcohol consumption. This study was conducted in the Department of Life Sciences - College of Science \University of Al-Qadisiya in cooperation with the laboratories of Al-Diwaniyah Teaching Hospital and the Center for Genetic Blood Diseases in Al-Diwaniyah affiliated to the Diwaniyah Health Department and some private laboratories. The study included (60) patients by (30) patients with osteoporosis of both sexes. (9) males and (21) females, their ages ranged between 26 to 75 years, and they were compared with the healthy group, which included 30 persons, (15) males and (15) females, their ages also ranged from 20 to 70 years.

Patients were diagnosed as osteoporosis and controls as normal by measuring bone mineral density (BMD), using dual energy x-ray absorptiometry (DXA). In addition, serum osteocalcin measured by enzyme linked immuno sorbent assay (ELISA). this study aims to assess the serum osteocalcin levels in osteoporosis patients and compared with healthy persons.

introduction

Osteoporosis is the most prevalent skeletal disorder characterized by low bone mass, changes in the microscopic structure of bone, and increased bone fracture. Osteoporotic fractures most commonly happen in the wrist, hip, pelvis, and spine (Doosti et al,2013).

WHO defined osteoporosis when bone mineral density (BMD) measured by dual energy X-ray absorptiometry (DEXA) scan is less than -2.5 standard deviation below the mean value for young adults for same age and sex (T- score), and further classified it into osteopenia, osteoporosis and severe osteoporosis according to BMD grading. Osteoporosis is one of leading cause and risk factor for fracture (Kanis et al,2008). The ability to measure bone density by DEXA scan has been one of the most significant advances in the investigation and treatment monitoring of Osteoporosis. BMD correlates

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strongly with bone strength and accounts for 60% to 80% of bone strength. Bone strength depends not only on the amount of mineral but also on the structural characteristics of the skeleton such as size, shape, and three-dimensional architecture (Stauber and Muller,2006; Ahlborg et al,2003). Until now the prediction of bone strength and risk of fracture has mainly been based on densitometry measurements. Lately, bone turnover biomarkers came into vogue to assess bone turnover rate and monitoring of treatment response in osteoporosis. Osteocalcin, also known as bone gamma-carboxyglutamic acid-containing protein (BGLAP) is secreted by osteoblasts during bone formation phase of bone remodeling (Hamdi,2013; Civitelli et al,2009). In osteoporosis, generally there is a deficiency of calcium and phosphorus level and since osteocalcin is a calcium dependent biomarker and has a strong affinity with bone matrix (hydroxyapatite) responsible for mineralization of bone. Osteoporosis leads to decreased hydroxyapatite crystal formation and hence results in increase in serum osteocalcin levels (Jagtap et al,2013; Filip et al,2004).

Materials and Methods

A case-control study was conducted between October 2020 and March 2021 and included 30 Iraqi patients from Al-Diwaniyah governorate suffering from osteoporosis and 30 healthy people, and it was conducted at Al-Diwaniyah Teaching Hospital and some private laboratories, and all patients and observers were from the same (Arab) ethnic group.

The subjects of the study consisted of 30 patients with osteoporosis who were selected from Al-Diwaniyah Teaching Hospital in their home (9) males and (21) females as an age group (26-75 years). The study of the control group included 30 people who appear to be in good health and they were both males and females in the age group (10-70) years. All subjects in this study obtained written consent before participating in this study.

They were subjected to BMD assessment by DEXA scan of lumbar spine. Those with t-score above -1 SD were taken as controls and with t-scores less than -1 SD were taken as cases. Bone mineral density was measured at the lumbar spine L1–L4 by using DEXA machine type (STRATOS, DMS, made in France). The results of measurement categorized according to WHO definition guidelines Normal: T-score (≥ -1.0); Osteopenia: T-score between (-1.0 and -2.5); Osteoporosis: T-score (≤ -2.5) (Cosman et al,2014). In addition, serum osteocalcin measured by enzyme linked immuno sorbent assay (ELISA).

The results were subjected to statistical analysis in order to find out the significant differences between the scores of the studied criteria and the comparisons were made using one-way analysis of variance (ANOVA1). The significant differences were determined at the level (0.05). All statistical analyzes were extracted using the program (spss10). The significant differences between the means were also tested using the least significant difference test (LSD) at a probability level of 0.05.

Results and discussion

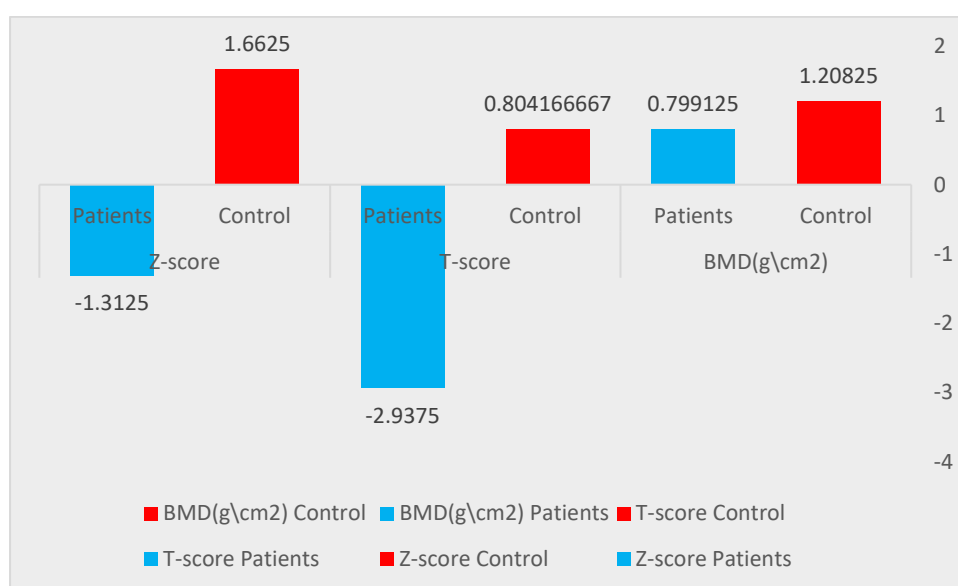
The mean age of participant was (mean \pm SD,55.35 \pm 3.14) of the Patients and (51.76 \pm 10.7) of the Controls.

It was found that the average degree of bone mineral density (BMD) in patients with osteoporosis decreased significantly compared to the control group as shown in (Table1) and (Figure1). Results of (Neetakumar et al., 2004) supported the results of our study.

The study also showed a significant reduction in T-score and Z-score in osteoporosis patients compared to healthy persons as described in (table 1) and (figure 1).

Table 1. Baseline characteristics of the studied patients compared to healthy persons.

Characteristics	Patients (Mean±SD)	Controls (Mean±SD)
Age	55.35±3.14	51.76±10.7
BMD	1.208 ±0.022	1.208±0.022
T-score Lumber spine	-2.93 ±0.001	0.804±0.004
Z-score	-1.312 ±0.002	1.662±0.041
Osteocalcin	47.95 ±0.53	38.53±2.68



(Figure 1) the values of BMD, score-T and score-Z in patients with osteoporosis compared to the control group.

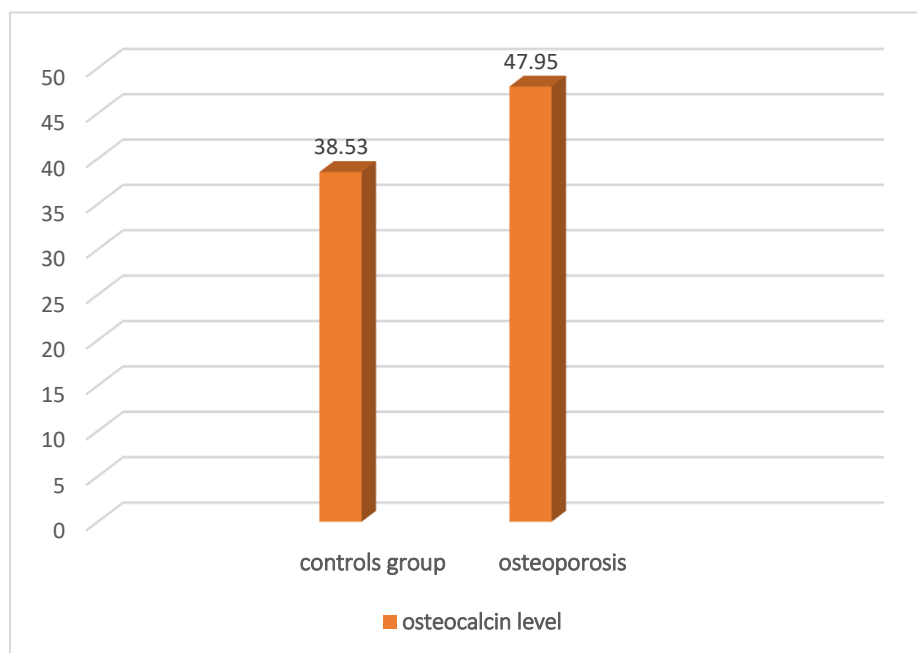
Where it was found that twenty patients suffer from osteoporosis (T-score $\leq -2.5SD$) in the rate 66.66% and ten patients had osteopenia (T-score between -1 and -2.5 SD) and a rate of 33.33% as shown in (Table 2).

Table (2) Dividing patients into two categories according to the T-score, the number of cases and the percentage of each category

Categories	T-score	No	Percentage
Osteopenia	T-score between -1 and -2.5 SD	10	33.33%
Osteoporosis	T-score $\leq -2.5SD$	20	66.66%

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The results of the study show high levels of serum osteocalcin in osteoporosis patients compared with healthy persons as shown in (Table 1) and (figure 2)



(Figure 2) The difference in osteocalcin values in osteoporosis patients compared to the control group.

This demonstrates an inverse relationship between serum osteocalcin level and bone mineral density in the current study. The inverse association between osteocalcin and BMD has been well documented in the literature (Filip, 2004; Bhattarai et al., 2014; Camara et al., 2014).

Osteocalcin is synthesized in the skeleton by osteoblasts, the cells responsible for bone formation. Osteocalcin is a major and most well-characterized non-collagen protein in mature human bone. It is a very sensitive scientist to bone formation. The advantage of using osteocalcin as a clinical indicator of bone turnover is its tissue specificity and is relatively low in person variance (Power and Fottrell, 1991).

An elevated level of osteocalcin may be associated with increased osteoblast activity. Osteocalcin has a high affinity for calcium and exhibits a compacted calcium dependent α -helix, whereby carboxy-glutamic acid (Gla) residues bind and enhance the uptake of hydroxyapatite into the bone matrix, in this way bone mineralization occurs. In women with osteoporosis, a deficiency of Calcium and phosphorous decrease the formation of hydroxyapatite crystals. Thus, if bone mineralization is low, free osteocalcin may be available for circulation in the blood. This may explain the increased concentration of osteocalcin in the blood serum of patients with osteoporosis. Examination of a specific marker of bone tumor activity, such as osteocalcin, reveals the predictive importance of osteocalcin for improving PMO management. (Pino et al., 1991) found osteocalcin to be a promising marker of bone turnover useful in the diagnosis and follow-up of elevated osteoporosis. Similar observations have been reported by a number of other studies (Verit et al., 2006; Cabrera et al., 1998; Rosenquist et al., 1995). Osteocalcin may be involved in the recruitment of osteoclasts to new bone sites and thus may function as a negative regulator (Ram et al., 2015). The problem, osteocalcin can be used as a marker of bone formation to give an idea of the change in bone turnover when it is normalized with respect to resorption (Becerik et

al., 2011). Osteocalcin plays a role in bone resorption and the mineralization process, and participates in the recruitment of osteoclasts to bone resorption sites and their formation so that they act as a negative regulator (Ram et al., 2015).

Conclusion

Measurement of osteocalcin is accessible, inexpensive and easy. Serum osteocalcin level can be used as a screening tool in hospitals to detect about osteoporosis.

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