eJBio

Electronic Journal of Biology, 2016, Vol.12(1): 8-11

A Study of Some Immunological and Biochemical Indicators for Patients with Osteoporosis

Laith B Alhusseini[1], Ali S Kadhim[2], Maytham M Alabassi[1], BA Almayahi[1,*]

1 Department of Environment, College of Science, University of Kufa, Najaf, Iraq;
2 Department of Science, Basic Education College, Waist University, Kut, Iraq.

*Corresponding author. 009647823004353; E-mail: basimnajaf@yahoo.com

Citation: Almayahi BA, Alhusseini LB, Kadhim AS, et al., A Study of Some Immunological and Biochemical Indicators for Patients with Osteoporosis. Electronic J Biol, 12:1

Received: December 22, 2015; **Accepted:** January 05, 2016; **Published:** January 11, 2016

Research Article

Abstract

In this study, focused on effect immune cells on bone and directed effect on osteoblast and osteoclast, which are secreted from cytokines and C-reactive protein and express trigger to immune response to continue to cause osteoporosis. Role of humoral immune response to osteoporosis is demonstrated. Some immunological indicators are studies for Iraqi patients. Blood samples are collected from all people (patients and healthy controls) to determine calcium levels, alkaline phosphatase enzyme, Erythrocyte Sedimentation Rate (ESR), Immunoglobulin G (IgG), and Immunoglobulin M (IgM). Results showed an increase in erythrocyte sedimentation rate (48.45) compared with healthy controls (16) as the value of T=16.89. There is a significant difference between patients and healthy controls $p<0.001$. The high rate of IgG (++) compared with IgM (+). It concludes that osteoporosis has a role in immune diseases and chronic diseases.

Keywords: Osteoimmunology; Immune system; Osteoclast calcium; Alkline phosphatase; ESR; IgG; IgM.

1. Introduction

Calcium is an essential mineral required by nearly every living organism. It is perpetually criminated from the body of skin cells, hair, nails, urination, sweat, and feces. If not replenished at adequate rates, the body will begin to scavenge the calcium it needs from the skeletal. Extensive studies on cross talk between immune and skeletal system in autoimmune disease give to rise to a new discipline of Osteoimmunology. The term "Osteoimmunology" used by Aaron Choi in the year 2000 the focus was on the relationship between the immune and skeletal system, especially in autoimmune diseases and other inflammatory [1]. The major discoveries and advances in this field led to the detection of molecular mechanisms as well as various cytokines and transducers refers to participate in the regulatory interaction between immune cells and bone cells. In addition, along with an arsenal of mutual signaling molecules, immune cells and bones also share a common site of origin, which is the bone marrow. Due to its proximity to the spatial development of the cells it suggests that they affect each other, not only after maturation and activation, as Kong and his colleagues already observed in 1999 [2,3], also at the beginning of its existence. Taichman and Emerson described the important role of osteoblasts in establishing outlets for hematopoietic stem cells, as well as in erosiontment and maintenance of Hematopoietic Stem Cells (HSC) in the bone marrow [4-8]. The development of bone cells has also been demonstrated to be supported by cells of the immune system; for instance, T cells are capable of influencing Osteoclastogenesis by the secretion of various cytokines such as interleukin-1, interleukin-6, interferon-γ or interleukin-4 [9,10], arose ideals Osteo immunological most prominent of note bone loss by osteoclast in various inflammatory diseases and autoimmune such as rheumatoid arthritis, diabetes, and lupus Erythematosus, and gum disease and viral infections, chronic (HIV) [2,11-15]. Kong et al. noticed that the carrying capacity of osteoclast activated T cells in rheumatoid arthritis Assistant brokered through activated RANKL/nuclear factor receptor κB (arrangement)/osteoprotegerin (OPG) axis [2]. RANKL is the main organizer for osteoclastogenesis, which contributes significantly to the process called bone remodeling. In this process, eating absorber old and damaged bone, which is replaced by new bone material deposited by osteoblasts? As normal physiological formation of bone remodeling is imperative to maintain bone strength, integrity, and imbalances lead to either increase or decrease bone mass of the latter it is often caused by inflammatory diseases. Cytokine regulating is the formation of osteoclast caused by lack of estrogen. Caused by estrogen deficiency bone loss is a complex effect of a large number of redundant passages of cytokines and work in a

cooperative manner to regulate osteoclastogenesis. Causing a lack of estrogen in a global increase in IL-7 production, especially in the thymus, and bone marrow and spleen. The increase in the production of IL-7 in the bone marrow increases T cell population, which increases the activity of TNF production and import from blood cell progenitor's origin mulberry in the thymus. The increase in IL-7 in the thymus gland increases production of the naive CD4+ T cells in peripheral blood, which in turn expansion of T cells, which gathered led to increases production of TNF. As estrogen deficiency independently or cooperatively increases in the production of tissue cells cytokines such as IL-6, IL-1, RANKL and M-CSF, which promotes the proliferation and differentiation osteoclast precursors to the bone marrow [16]. This paper focused on study immunological response and effect it on bone mass during some changing immunological (ESR, IGg, and IGM) and biochemical (BAP and Ca) indicators for patients with osteoporosis on January to December 2014.

2. Materials and Method

100 samples were collected from people suffering from joint and bones pains, fatigue and debilitated. Where it was diagnosed 55 of the sample infected with osteoporosis. The study of the disease is not specific to sex, but in general, by taking random sample for the study of the disease immunologically for both sexes. 55 samples of blood collected from patients of osteoporosis (male and female) aged (35-75 years) from Al Kindi General Teaching Hospital in Baghdad city on January to December 2014. Also, 50 samples of blood collected from healthy controls. After conducting laboratories tests and clinical diagnosis by doctors' competence in the field of joints and fractures, blood samples from the patient of 5 ml were collected from the blood of a vein from the arm. The samples are left for 15 minutes at room temperature where it was clotting. Tube of blood samples is placed in a centrifuge at speed (3000) rpm for 15 minutes and then blood serum is isolated. The blood samples are collected in the same time from the patients (fasting) in the morning, the patient did not give any medication (before treatment). Various tests conducted on the sample in the same day. It was measured calcium ion concentration (Ca++) by colorimetric methods [17]. Also, it was measured BAP enzyme secretion of patients and healthy controls in the same method [18]. The speed of erythrocyte sedimentation rates measurement for their immunological tests by traditional method (Westergren method) and Antibody plus Combo Rapid Test (APCRT) are used [19]. Immunological antibodies titer based on the principle of simple reaction (AB-AG REACTION) have been measured in the serum of patients and healthy controls. Effective protein titer rate depending on the CRP-index agglutination titter method have been measured [20]. Statistical analyses of data were conducted using SPSS software (2012) for Windows. Equality of several means was tested using one-way classification. The data were analyzed using T test. Various statistical analyses were performed for the data. Many published statistical analyses quote p-values as ≥ 0.05 (no significant), <0.05 (significant), and <0.01 (highly significant) [21].

3. Results and Discussion

Table 1 show that the percentage of men has the disease in osteoporosis (bone necrosis), it was (23.6%) for men compared to women whose percentage (76.4%) as the samples collected after diagnosis ages ranging from 40 y to 60 y. The results showed that women are more susceptible to osteoporosis than men, especially women with postmenopausal for the collected samples. In this study Nelson et al. and Olszynski et al. [22,23] showed that osteoporosis in women at postmenopausal are more than men in Canada.

Table 1. Osteoporosis patients.

Sample type	Number	Total Number	Percentage
Male	13	55	23.6
Female	42		76.4

Table 2 showed that low calcium rate in patients with osteoporosis (8.28) compared with healthy controls, where the rate of calcium (10.16) which is within normal limits (T=11.199) and there is statistically significant (p<0.001). This means that calcium levels in the blood of patients goes down, so the hardness of bones is few. Where, calcium is a key element in myeloid mass components [24]. Arnaud and Sanchez [25] showed the reason for the low rate of calcium ratio of patients to an imbalance in the absorption of calcium from food as a result of hormonal imbalance. Then renal function less in production 1.25 $(OH)_2D_3$, which has a role in the body's fluid balance (electrolytic) in the blood then the body begins to withdraw calcium from the bone marrow by an estimated (1500-2500 mg/day). This

Table 2. The concentration average of calcium.

Sample type	Average (mg/dl)	T-Test	Sig(2-taild)
Patients	8.2891	11.199	0.000
Controls	10.1655		

amount is difficult to compensate and therefore osteoporosis occur.

Table 3 shows the increase in enzyme secretory for patients at a rate of (84.26) compared with healthy controls, where it was observed that the enzyme secretory within normal limits (40.38) at T=14.606.

Table 3. Enzyme rate (BAP).

Sample type	Average (IU/l)	T-Test	Sig(2-taild)
Patients	84.2636	14.606	0.000
Controls	40.3891		

There is a significant difference between patients and healthy controls p<0.001. These results indicate an imbalance in the cellular secretory of this enzyme located on the cell surface. Kress [26] showed an imbalance in the secretion of the cells of the bone, which increases the secretion of this enzyme in order to keep the basal case that must be available for the continuity of calcium to build bone.

Results showed an increase in erythrocyte sedimentation rate (48.45) compared with healthy controls (16.0) as the value of T=16.89. There is a significant difference between patients and healthy controls p<0.001 as shown in Table 4. The high

Table 4. Red blood cell sedimentation rate.

Sample type	Average (mm/hr)	T-Test	Sig(2-taild)
Patients	48.45	16.892	0.000
Controls	16.00		

erythrocyte sedimentation rate is an indication of the presence of inflammation in the body and stimulates the immune system. Pisetsky [27] explained that the examination of the ESR increases in inflammatory conditions of autoimmune diseases and inflammation of the tonsils and bone inflammation and cancer.

Table 5 shows the high rate of IgG (++) compared

Table 5. Antibody titer rate (IgM, IgG).

Sample type	IgM	IgG
Patients	+	++
Controls	NEGATIVE	NEGATIVE

with IgM (+). It concludes that osteoporosis has a role in immune diseases and chronic diseases.

Table 6 illustrates the high effective rate of protein in patients (1094.73) compared with healthy controls (4), as the value of T=15.955. There is a significant

Table 6. Effective protein level (CRP).

Sample type	Average (mg/L)	T-Test	Sig (2-taild)
Patients	1094.7368	15.955	0 000
Controls	4		

difference between patients and healthy controls p<0.001. Where, protein is a means of inflammatory and defense produced by cells during inflammation (the immune attack) as well as secreted by the liver as an indicator of the occurrence of an immune attack [28,29]. Results show the role of the immune system immune inflammatory such as TNF, RF, CRP, and IL1-2-6 in the production of inflammation in the bone tissue and the occurrence of bone necrosis as shown in Tables 4-6. Goldering and Siggelkow et al. [30,31] showed the existence of a biological overlap between bone disease and inflammation of the immune. Manolagas [32] showed that Cytokines have a key role in osteoporosis. Where, IL 6 works on creating, activating, and stimulating the cells of the bones cationic Osteoclast where secreted interleukin of immune cells (neutrophilis and monocytes), which is one of the pro-immunity cells. The role of interleukin with so much of bone diseases such as Patyets disease and osteoporosis. Wei et al. [33] explained that interleukin has a role in reducing the absorption of bone for mineral materials for building bone material and thus works on osteoporosis.

4. Conclusion

Low calcium rate in patients with osteoporosis (8.28) compared with healthy controls. The rate of calcium (10.16) within normal limits (T=11.199). It was observed that the enzyme secretory within normal limits (40.38) at T=14.606. There is a significant difference between patients and healthy controls p<0.001. These results indicate an imbalance in the cellular secretory of this enzyme located on the cell surface. Effective rate of protein in patients (1094.73) found to be high compared with healthy controls (4), as the value of T=15.955. There is a significant difference between patients and healthy controls p<0.001.

Reference

[1] Aaron J, Choi Y. (2000). Bone versus immune system. *Nature*. **408**: 535-536.

[2] Kong YY, Feige U, Sarosi I, et al. (1999). Activated T cells regulate bone loss and joint destruction in adjuvant arthritis through osteoprotegerin ligand. *Nature*. **402**: 304-309.

[3] Rifas L, Arackal S, Weitzmann MN. (2003). *J Cell Biochem*. **88**: 650-659.

[4] Dazzi F, Ramasamy R, Glennie S, Jones SP, Roberts

I. (2006). Activated T cells regulate bone loss and joint destruction in adjuvant arthritis through osteoprotegerin ligand. *Blood Rev*. **20**: 161-171.

[5] Jung Y, Wang J, Havens A, et al. (2005). Cell-to-cell contact is critical for the survival of hematopoietic progenitor cells on osteoblasts. *Cytokine*. **32**: 155–162.

[6] Neiva K, Sun YX, Taichman RS. (2005). The role of osteoblasts in regulating hematopoietic stem cell activity and tumor metastasis. *Braz J Med Biol Res*. **38**: 1449-1454.

[7] Taichman S, Emerson G. (1998). The role of osteoblasts in the hematopoietic micro environment. *Stem Cells*. **16**: 7-15.

[8] Taichman RS, Reilly MJ, Emerson SG. (2000). The role of osteoblasts in the hematopoietic micro environment. *Hematology*. **4**: 421-426.

[9] Mirosavljevic D, Quinn JM, Elliott J, et al. (2003). T-cells mediate an inhibitory effect of interleukin-4 on osteoclastogenesis. *J Bone Miner Res*. **18**: 984-993.

[10] Takayanagi H, Ogasawara K, Hida S, et al. (2000). T-cell-mediated regulation of osteoclastogenesis by signaling cross-talk between RANKL and IFN-gamma. *Nature*. **408**: 600-605.

[11] Konishi M, Takahashi K, Yoshimoto E, et al. (2005). Association between osteopenia/osteoporosis and the serum RANKL in HIV-infected patients. *AIDS*. **19**: 2040-2041.

[12] Kotake S, Udagawa N, Hakoda M, et al. (2001). Activated human T cells directly induce osteoclastogenesis from human monocytes: possible role of T cells in bone destruction in rheumatoid arthritis patients. *Arthritis Rheum*. **44**: 1003-1012.

[13] Shigeyama Y, Pap T, Kunzler P, et al. (2000). Expression of osteoclast differentiation factor in rheumatoid arthritis. *Arthritis Rheum*. **43**: 2523-2530.

[14] Stellon AJ, Davies A, Compston J, Williams R. (1985). Bone loss in autoimmune chronic active hepatitis on maintenance corticosteroid therapy. *Gastroenterology*. **89**: 1078-1083.

[15] Mundy GR. (2002). Metastasis to bone: causes, consequences and therapeutic opportunities. *Nat Rev Cancer*. **2**: 584-593.

[16] Renqing Z. (2012). Immune Regulation of Osteoclast Function in Postmenopausal Osteoporosis: A Critical Interdisciplinary Perspective. *Int J Med Sci*. **9**: 825-832.

[17] Tietz N W. (1971). Clinical guide to laboratory tests, 3rd edition. W.B. Saunders co. Philadelphia, PA.

[18] Belfield A, Goldberg DM. (1971). Revised assay for serum phenylphosohatase activity using 4-amino-antipyrine enzyme. *Enzyme*. **12**: 561-573.

[19] Dawood MY, *Saxeba BB, Landesman R, Ob. Gyn.* (1990). 126-678.

[20] Hason L. (1997). *Current opinion in factious disease*. **10**: 196-201.

[21] Almayahi BA. (2013). Study on radioactivity in human teeth, animal bones and soil in selected areas in northern region of Malaysian peninsular. *Universiti Sains Malaysia*.

[22] Nelson HD, Helfand M, Woolf SH, Allan JD. (2002). Screening for postmenopausal osteoporosis: a review of the evidence for the U.S. Preventive Services Task Force. *Ann Intern Med*. **137**: 529-541.

[23] Olszynski WP, Brown JP, Adachi JD et al. (2014). Normative data for multisite quantitative ultrasound: the Canadian Multicenter Osteoporosis Study. *J Clin Densitom*. **17**: 534-40.

[24] Arnaud CD, Sanchez SD. (1990). The role of calcium in osteoporosis. Annual Review of Nutrition. *Annual Review of Nutrition*. **10:** 397- 414.

[25] Arthur JV, James HS. (2001). Human physiology, the mechanisms of body function, 13th edition, McGraw – Hill international.

[26] Kress BC. (1998). Bone alkaline phosphatase: methods of Quantition and clinical utility. *J clin ligand assay*. **21:** 139–148.

[27] Pisetsky DS. (2013). Medline plus. National library and national institutes of health, retrieved 8 July 2013.

[28] Muller B. (2002). Cytokine imbalance in non-immunological chronic disease Cytokine. *Cytokine*. **18:** 334-339.

[29] Koh JM, Kang YH, Jung CH et al. (2005). Higher circulating hsCRP levels are associated with lower bone mineral density in healthy pre-and postmenopausal women: evidence for a link between systemic inflammation and osteoporosis. *Osteoporosis Int*.

[30] Goldring SR. (2003). Inflammatory mediators as essential elements in bone remodeling. *Calcif Tissue Int*. **73:** 97-100.

[31] Siggelkow H, Eidner T, Lehmann G, et al. (2003). Cytokines, osteoprotegerin, and RANKL in vitro and histomorphometric indices of bone turnover in patients with different bone diseases. *J Bone Miner Res.* **18:** 529-38.

[32] Manolagas SC. (2000). Cytokines, osteoprotegerin, and RANKL in vitro and histomorphometric indices of bone turnover in patients with different bone diseases. *Endocr Rev.* **21:** 115-137.

[33] Wei S, Kitaura H, Zhou P, Ross FP. (2005). IL-1 mediates TNF-induced osteoclastogenesis. *J Clin Invest*. **115:** 282-90.