Relation between Hyperparathyroidism and Osteoporosis in Chronic Renal Failure Patients with Regular Haemodialysis

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Abstract: The aim of this study was to find a relation between hyperparathyroidism and osteoporosis in chronic renal failure patients (CRF) with regular haemodialysis (HD) at haemodialysis unit of Al-Hussein University Hospital. 100 CRF patients on regular HD and 20 healthy controls were studied. All included patients and controls were subjected to the following: Full history taking, complete clinical examination, complete blood picture, blood urea, serum creatinine, serum calcium, serum phosphorous, Aspartate transaminase (AST), Alanine transaminase (ALT), serum albumin, serum alkaline phosphatase (AP), serum iron, serum ferritin and total iron" binding capacity. Serum intact parathyroid hormone (iPTH) by immunoradiometricassay (IRMA). Bone density by quantitative heel ultrasound (QUS). The results of this study revealed that 27% of haemodialysed patients had normal bone mineral density (HMD) (T-score above-1), 53% were osteopenic (T-score between-1 to-2.5) and 20% showed Osteoporosis (T-score less than -2.5). There were significant increase in serum iPTH levels in 40% of our haemodialysed patients, these results indicate that reduced BMD was due to osteopenia and Osteoporosis in addition to renal osteodystrophy as indicated by elevated serum: iPTH in 40% of our patients. These results indicate that 33% of our patients had frank osteopenia and Osteoporosis other than renal osteodystrophy. There were significant negative correlation" between BMD and iPTH, serum phosphorous, age, body weight, body mass index and duration of haemodialysis. There were significant positive correlation between BMD and serum calcium, height and sex as incidence was more in females. In conclusion: osteopenia and Osteoporosis among patients undergoing haemodialysis were common..Static and kinetic bone histomorphometry after double tetracycline labelling and bone biopsy are essential for assessment of the bone disease in uremic patients especially in high risk or recurrent fractures.
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1.Introduction

Chronic kidney disease (CKD) patients have an increase risk fractures ***(Kidney disease improving clinical outcomes, 2009).***

Patients with mineral and bone disorders seen with CKD (CKD-MBD) have complex abnormalities in bone physiology ***(Ott et al., 2009).***

Osteoporosis is a disease of bone that leads to an increased risk of fracture. In Osteoporosis the bone mineral density (BMD) is reduced, bone microarchitecture is disrupted, and the amount and variety of non-collagenous proteins in bone is altered (WHO, 1994), The underlying mechanism in all cases of Osteoporosis is an imbalance between bone resorption and bone formation (Raisz, 2005). Disturbances in mineral and bone metabolism are common in patients with chronic' kidney disease (CKD). End-stage renal disease (ESRD) patients usually have accelerated bone loss' due to abnormal boneturnover that leads to high prevalence of bone health problems, e.g., osteopenia and osteoporosis, furthermore, parathyroid hormone (PTH)-related bone disease influences bone mineral density in hemodialysis patients, in addition to other important risk factors such as advanced age, age at menarche, female gender, and history of previous fractures, etc. On the other hand, risk factors for bone mass loss in this population include body weight, hemoglobin and weekly heparin dose (Gal-Moscovici and Sprague, 2007). Huang, *et al.* (2009) reported that 81% of hemodialysed patients had a T-score lower than -1, of them 13% had osteoporosis with the femoral neck most commonly affected. Regarding risk factors, age, serum alkaline phosphatase (ALP) level and intact parathyroid hormone had significant negative correlations with the femoral neck bone mineral density, also low body weight and low serum albumin level were associated with a low bone mass in the hemodialysis patients. The diagnosis of osteoporosis in postmenopausal women is based on bone mineral density criteria established in 1994 by The World Health Organization (WHO; T score of -

2.5 or lower) or the presence of the fragility fractures (WHO Study Group, 1994). However, these criteria cannot be used to diagnose osteoporosis in the patient with CKD or end-stage renal disease (ESRD) because all of the various forms of renal osteodystrophy that are not osteoporosis also have low T scores and may develop fragility fractures (Cunningham, *et al.*, 2004). The only way to make the diagnosis of osteoporosis in patients with CKD or ESRD is by excluding the other forms of renal osteodystrophy. This can be done to some degree by biochemical profiling, measuring in particular the parathyroid hormone (PTH) level and the bone specific alkaline phosphatase (BSAP) (Elder, 2002). The aim of this study was to find a relation between hyperparathyroidism and osteoporosis in chronic renal failure patents on regular haemodialysis.

This study was conducted on 100 chronic renal failure (CRF) patients on regular hemodialysis (HD) in nephrology and dialysis unit at AL-Hussien University Hospital (group 1), in addition to 20 normal control subjects (group 2).

Group 1:

Included 100 patients with CRF on regular HD for at least 6 months, 51 of them were males and 49 were females, their ages ranged between 26 and 62 years with a mean value of 49.37 ± 1.02 years. The duration of HD ranged between 1-16 years with a mean value of 4.2±1.3. During dialysis the patient's received heparinization with 10000 units as a maximum dose. Erythropioetin was given to each patient according to body weight (100-150 lu/kg/week), alfa calcidol in a dose of 0.5(ig/day combined with 1500-3000mg elemental calcium/day. Dialysis was performed for 4 hours three times weekly. Blood access was through arterio-venous fistula. Blood flow rate was usually 300-350ml/min with a dialysate flow rate of 500ml/min. Ultrafiltration varied according to patient's actual weight. The membrane used was the Fresenius polysulfone 1.3 and 1.7 models, with surface area suitable for each patient. Bicarbonate was the buffer used throughout the study for all patients.

Group 11:

Included 20 healthy subjects as controls with normal renal function, 11 of them were males and 9 were females, their ages ranged between 26 and 60 years with a mean value of 47.4 ± 2.64 years. All patients and controls were subjected to the following:

Detailed history and clinical examination-Body mass index (BMI) was calculated according to Pi-Sunyer (1998). Laboratory investigations included: -Complete blood count, blood urea, serum creatinine, serum calcium, serum phosphorous, serum alanine transaminase (ALT), and aspartate transaminase (AST) enzymes, serum albumin and serum alkaline phosphatase. Serum intact parathyroid hormone (iPTH) by immunoradiometricassay (IRMA) (Gerakis, *et al.*, 1996). Serum iron estimated by the colorimetric method (Callahan and Cook, 1982). Serum ferritin using ELISA coated microtitre strips (Wick, *et al.*, 1996). Total iron binding capacity (TIBC) by automated analyzer (Siek, *et al.*, 2002).

Bone density by ultrasound:

Quantitative heal ultrasound (QUS) is a mobile, relatively inexpensive, easy to perform and radiation-free method which can predict fractures to the same extent as dual-energy X-ray absorpiometry (DXA). Quantitative heal ultrasound methods are promising tools for the assessment of the bone status in osteoporosis. The techniques are based on changes in speed and amplitude of a broad band ultrasound signal propagating through the calcaneous bone. The result by T-score which indicate T-Score (normal-1.0-2.0), osteopenia less than-1.0 to-2.5, osteoporosis less than-2.5 to-4.0 (Lindberg and Moe, 1999).

Statistical analysis:

Data were analyzed using SPSS program version 13. Results were expressed as mean ± SD for normally distributed .data, and percentages for categorical data. Comparison between two groups was carried out with the student (t) test. In correlation studies data were analyzed using Pearson's bivariate correlation with regression equation, and the degree of association was expressed as ® correlation coefficient and results were reported by probability values. Values of $P<0.05$ were taken as statistically significant.

Results:

Clinical and biochemical parameters, as well as significant differences between studied groups were presented in table 1. There were no significant differences between control subjects and haemodialysed(HD) patients regarding body weight, height, body mass index, serum ALT, serum AST and serum iron. There were significant difference between control subjects and HD patients regarding age. Hemoglobin, serum calcium, serum albumin, serum alkaline phosphatase, TIBC andbone density were significantly lower in HD patients when compared to controls, (P <.005, P <.02, P<.04, P <.005, P <.005, P <.005) respectively. Blood urea, serum creatinine, serum phosphorous, serum ferritin and serum parathyroid hormone (iPTH) were significantly higher in HD patients than controls, (P<.005, P<.005, P<.009, P<.005, P <.02) respectively.

Correlations between bone density (T-score) and clinico-biochemical parameters of HD patients were presented in table 2. There were significant

negative correlation between bone density and age of HD patients (r=-0.26, *P* <.008), body weight (r=-0.24, *P* <.01), body mass index (r=-0.21, *P* =0.03), duration of haemodialysis (r=-0.21, *P* =0.03), serum phosphorous (r=-0.20, *P* = 0.01), and serum iPTH (r=-0.35, *P* <.005). There were significant positive correlation between bone density and sex (r=0.47, *P* <.005), height (r=0.31, *P* <.005), and serum calcium (r=0.20, *P* <0.04).

Table (1): Comparison between clinical and biochemical parameters for studied groups.

Groups / Parameters	Group 1 (HD) patients (n=100)		Group 2 (normal controls) (n=20)		t	*P*
	Range	Mean ± SD	Range	Mean ± SD	3.45	<0.003
Age (years)	26-62	49.37±1.02	26-60	47.4±2.64		
Body weight (kg)	40-95	64.34±1.28	51-76	62.4±1.68	1.60	0.125
Height (meter)	1.53-1.76	1.61±0.004	1.52-1.73	1.63±0.01	-0.685	0.50
Body mass index (kg/m2)	16.02-24.59	24.95±0.44	20.17-27.91	23.6+0.41	2.43	0.06
Hemoglobin (g/dl)	6.4-14	9.6±0.18	11.9-15	13.6±0.17	-7.2 '	<0.005
Blood urea (mg/dl)	58-318	146±54.56	21-38	29.65±1.12	10.28	<0.005
Serum creatinine (mg/dl)	3.4-17.2	9.5+0.27	0.5-1.4	1+0.05	14.48	<0.005
Serum calcium (mg/dl)	3.8-13	8.89±0.182	9.3-10.7	9.97+0.08	-2.45	<0.02
Serum phosphorous (mg/dl)	1.1-9.8	5±0.18	3.4-4.3	3.99±0.06	2.93	<0.009
Serum ALT (U/L	3-742	32±7.49	16-37	26.35±1.44	-0.62	0.53
Serum AST "(U/L)	4-172	29.9±2.5	14-34	24.55±1.37	0.441	0.66
Serum albumin (g/dL)	2.8-4.9	3.93±0.03	3.6-4.9	4.2+0.08	-2.18	<0.04
Serum ALP (U/L)	3.4-194	13.7±2.33	49-65	57.9±0.98	-17.63	<0.005
Serum iron (g/dL)	22-251	111.51+538	82-135	109±3.85	0.04	0.96
Serum ferritin (ng/mL)	15-8558	1542.1±5382	65-107	82.3±2.55	4.87	<0.005
TIBC (g/dL)	117-345	215.6±4.26	270-395	324±7.2	-9.76	<0.005
Serum iPTH (pg/mL)	14-2189	280.5136.1	23-47	35.55±X.59	2.44	<0.02
Bone density (T -score)	-4.7-2.3	-1.66±0.12	-0.6-1	0.25±0.11	-6.46	<0.005

ALT = Alanine transaminase, AST = Aspartate transaminase, ALP = Alkaline phophatase, TIBC = Total iron binding capacity, iPTH = intact parathyroid hormone.

Table (2): Correlation between bone density (T-Score) and clinical and biochemical parameters of hemodialysed patients.

Parameters	r	*P*	Significance
Age	-0.26**	0.008	H.S.
Sex	0.47**	<0.005	H.S.
Body weight	-0.24	0.01	S
Height	0.31**	<0.005	H.S
Body mass index	-0.21*	0.03	S
Duration of Hemodialysis HHHDDhemodialysis	-0.21*	0.03	S
Serum calcium	0.20*	0.04	S
Serum phosphorous	-0.20*	0.01	S
Serum ferritin	-0.22	0.013	NS
Serum iPTH	-0.35**	<0.005	HS

S=Significant*, H.S = highly significant**, NS = nonsignificant.

4. Discussion

Disturbances in mineral and metabolism are common in, patients with chronic kidney disease (CKI).These patients have bone pain, increased incidence of bone fractures and deformity, myopathy and muscle pain, and ruptures of tendons. Hyperphosphatemia also appears to be associated with increased mortality, and elevatedblood levelsof PTH exert significant adverse effects on the function of almost every organ (Miller, 2003).

The present study was designed to showed a relation between hyperparathyroidism and osteoporosis and related biomarkers that leads to chanes in bone density in HD patients. The present study revealed that 27% of our hemodialysed patients had normal bone mineral density (BMD) (T-Score

above-1) whereas 53% were osteopenic (T-Score between - 1 to - 2.5) and 20% showed osteoporosis (T-score below-2.5). Huang, *et al.* (2009) found that 81 % of HD patients had a T-score lower than-1,13% of them had osteoporosis.

These results demonstrated that the bone density was reduced in HD patientsinspite of that they received alfacalcidol in a dose of 0.5 ug/day combined-with 150-3000mg elemental calcium/day. We assume that alfacalcidol use in dose of 0.5ug/day was not sufficient to protect bone of our patients. The explanation of these results may be due to impaired intestinal absorption of calcium as well as reduced action of vitamin D in those; patients. Also, reduction in available, calcium stimulates the release of parathyroid hormone leading to bone resorption.

The risk of bone loss and fractures varies with the age, menopausal status and risk factors related to the underlying disease (Sambrook, 2000). • The bone metabolism could be affected differently in patients with reduction in glomerular filtration rate (GFR) due to aging than it would be in patients with GFR reduction due to a specific renal disease (Klawansky, *et al.*, 2003). In our study, there were inverse correlation between age and bone density among HDpatients (group 1), and this agree with Klawansky, *et al.* (2003) and Melton, (2003), who found that, with age progression, there was decreased bone density. The risk of fracture was found to increase after the start of haemodialysis; the riskremained independent of underlying disease, age and gender (Van Staa *et al.*,2002).

In our study, we found that 73% of hemodialysed patients had a lower BMD and 40% had elevated iPTH levels, these results indicate that reduced BMD was due to osteopenia and osteoporosis, in addition to renal osteodystrophy as indicated by elevated serum iPTH in 40% of our patients. These results indicate that 33% of our patients had frank osteopenia and osteoporosis other than renal osteodystrophy.

In this work,' there were significant inverse correlation between body weight and bone density, and this agree with Ebeling (2008) who found that the.increase in body weight was considered a risk factor for osteoporosis, but this findings differe from the study of Hiiang *et al.* (2009) who stated that, low body weight and low serum albumin level were associated with a low bone mass in the hemodialysis patients. In this study, there were highly significant positive correlation between height and bone density. As regard body mass index, there were significant negative correlation between body mass index and bone density. These results were in agreement with Ebeling, (2008) who found that the increase in the body mass index predispose to osteoporosis. In the present work, there were significant negative correlation between duration of haemodialysis and bone density, and this can be explained by increased bone resorption and decreased bone formation in patients undergoing longterm haemodialysis which predispose to osteoporosis; Also another risk factor which may predispose to increase incidence of osteoporosis with increased duration of haemodialysis is the chronic acidosis. These results were in agreement with Block *et al.* (1998) who found that the increase in the duration of haemodialysis was associated with increased risk of osteoporosis.

The most important risk factors for osteoporosis are advanced age (in both men and women) and female sex, estrogen deficiency following menopause is correlated with rapid reduction in BMD, while in men a decrease in testosterone levels has a comparable (but less pronounced) effect (Melton, 2003). In the present work, we found that the osteoporosis was more prevalent among female patients, there were 12 out of 49 female patients (24.4%) had osteoporosis (T-score less than-2.5) in comparison to" 8 out of 51 male patients (15.9%), and this agree with Melton (2003) who found that osteoporosis was more common in female after menopause mostly due to estrogen deficiency.

These results brought to light the possible relationship between estrogen deficiency and changes in bone mass in haemodialysed women. These results suggest that estrogen status may play an important role in bone mass conservation in uremic women. Gonzalez *et al.* (2000) demonstrated that young amenorrhagic women on haemodialysis (<50years) showed significantly lower trabecular bone mineral density, lower estradiol levels, higher FSH levels, and increased bone resorption markers when compared with regularly menstruating women. A controlled study by Matuszkiewicz-Rowinsk *et al.* (1999) in a small group of pre-menopausal dialysis women showed that after 1 year .of transdermal estradiol with cyclic addition of nortestisterone acetate, the lumber spine bone mass density of treated women was significantly higher than that of untreated women.

Alkaline phosphatase(AP) is glycosylated protein produced by at least five different organs, namely. liver, bone, kidney, intestine and placenta. In uremia the relative contribution of each of these isoenzymes to serum total AP activity may be modified by changes in their relative half-lives (Schmidt-Grayak *et al.*, 1996). In bone, AP is produced by osteoblasts and osteoclast precursors and participates in the mineralization process. The development of monoclonal antibodies specific for bone alkaline phosphatase provided the basis for a

more specific index of bone formation (Hill and Wolfert, 1989). In this work, we found that the number of HD patients who have serum total AP below normal levels were 94 out of 100 patients, and 1 patient had AP above the normal value, while 5 patients have AP within the normal range.

These findings suggest that the reduced serum AP levels as a marker of bone formation in HD patients may act as a potential risk factor, at least in part, for the development of Osteoporosis in HD patients. Patients with CKD almost always develop secondary hyperplasia of the parathyroid 'glands, resulting in elevated blood levels of parathyroid hormone (PTH). This abnormality is due to the hypocalcemiathat develops during the course of kidney disease and/or to deficiency of 1.25-dihydroxycholecalciferol [1.25(OH)2D3] that may directly affect the function of the parathyroid glands (Hory and Drueke, 1997). In the present study, there were significant positive correlation between bone density and serum calcium, the explanation of these results could be due tohypocalcemia which increase synthesis of PTH which predispose to high bone turnover disease and decrease bone density. In our study the number of patients who had hypocalcemia were 33%, 17% of them had T-score less than-1.5.

Several lines of evidence suggest that phosphate retention can provoke secondary hyperparathyroidism, therefore, phosphate retention and hyperphophatemia can provoke secondary hyperparathyroidism in the absence or presence of impaired kidney function. As regard serum phosphorous, there were significant negative correlation between bone density and serum phosphorous, and this agree with Lock and Port, (2000) who reported that hyperphosphatemia occurs in a greater •proportion of patients on regular haemodialysis which have significant impact on patient's outcome.

In the present study, there was significant negative correlation between serum iPTH and serum calcium. These result was in agreement with Moallem, *et al.*(1998) who reported that a decrease in extracellular calcium concentration leads not only to an increase in iPTH secretion but also to increase in PTH mRNA levels and parathyroid proliferation.

Conclusion:

We can conclude that the osteopenia and Osteoporosis in haemodialysed patients were prevalent. It seems that alfacalcidole 0.5|u,g/day combined with calcium supplement was not sufficient, and it seems that antiresorptive therapies are warranted. Also, the diagnosis of Osteoporosis by using ultrasound was insufficient and double tetracycline-labeled quantitative bone histomorphometry and bone biopsy are essential for assessment of the bone disease in uremic patients especially in high risk patients or patients with recurrent fractures. Proper treatment of secondry hyperparathyroidism may be of benefit in decreasing the rate of fractures and also decreasing modifiable risk factors may do.

References

1. Block, G.A., Hulbert-Shearon, I.E., Levin, N.W. and Port, F.K (1998): Association of serum phosphorus and calcium-phosphate product with mortality risk in chronic hemodialysis patients: A national study. Am.J. Kidney Dis., 31:607-617.
2. Brown, A.J., Ritter, C.R., Urena, P., Finch, J.L. and Gagne, E. (1993) R.: A mechanism for the disparate actions of the calcitriol and 22-oxacalcitriol in the intestine. Endocrinology, 133: 1158-1164.
3. Callahan, J. and Cook, K. (1982): Serum iron estimation using colorimetric method. Anal. Chem., 54:59-61.
4. Cunningham, J., Sprague, S.M., Cannata-Andia, J., *et al.* (2004): Osteoporosis in chronic kidney disease.Am.J.KidneyDis,43:566-571.
5. Drueke, T.B. (1995): The pathogenesis of parathyroid gland hyperplasia in chronic renal failure. Kidney Int., 48:259-272.
6. Ebeling, PR. (2008): Clinical practice, osteoporosis in men. N. Engl. J. Med., 358(14):1474-1482.
7. Elder, G. (2002): Pathophysiology and recent advances in the management of renal osteodystrophy. Osteoporosis Int., 17(12): 2094-2105.
8. Gal-Moscovici, A. and Sprague, S.M. (2007): Osteoporosis in chronic Kidney disease, Semin. Dial., 20:423-430.
9. Gerakis, A., Hutchison, A.J., Apostolou, T., Freemont, A.J. and Billis, A. (1996): Biochemical markers for non-invasive diagnosisofhyperparathyroid bone disease and adynamic bone in patients on haemodialysis. Nephrol. Dial. Transplant., 11:2430-2438.
10. Gonzalez, L., Alvarez, H., Weisinger, J.R. *et al.* (2000): Role of persistantamenorrhea in bone mineral metabolism of young haemodialysed women. Kidney Int., 58:331-335.
11. Hill, C.S. and Wolfert, R.L. (1989): The preparation of monoclonal antibodies which react preferentially with human bone alkaline phosphatase and not liver alkaline phosphatase. Clin. Acta, 186: 315-320.
12. Hory, B. and Drueke, T.B. (1997): The

parathyroid-bone axis in uremia: New insights into old questions. Curr. Opin. Nephrol. Hypertens., 6:40-48.
13. Huang, G.S., Chu, T.S., Lou, M.F., Hwang, S.L. and Yang, R.S. (2009): Factors associated with low bone mass in the hemodialysis patients- a cross-Sectional correlation study. BMC Musculoskelet. Disord., 10:60-64.
14. Kidney Disease: Improving global outcomes CKDMBDWG. KDiGo clinical practice guideline for diagnosis evaluation, prevention and treatment of chronic kidney disease – Mineral and bone disorders (CKD-MBD), kidney Int. Suppl. 2009: 51-130.
15. Klawansky, S., Komaroff, E., Cavanaugh, P.P., Mitchell, D.Y. Gordon, M.J., Connelly, J.E. and Ross, S.D. (2003): Relationship between age, renal function and bone mineral density in the US population. Osteoporosis Int., 14:6848-6853.
16. Lindberg, J.S. and Moe, S.M. (1999): Osteoporosis in end-stage renal disease. Semin. Nephrol., 19:115-122.
17. Lock, G. and Port, F. (2000): Re-evaluation of risks associated with hyperphosphatemia and hyperparathyroidism in dialysis patients. Am.J. Kidney Dis,35: 1126- 1137.
18. Matuszkiewicz-Rowinska, Skorzewska, K., Radowicki, S. *et al.* (1999): The benefits of hormone replacement therapy in pre-menopausal women with estrogen deficiency on haemodialysis. Nephrol.Dial. Transplant, 14:1238-1243.
19. Melton, L.J. (2003): Epidemiology worldwide. Endocrinol. Metab.Clin: North Am.,32(1): 1-13.
20. Miller, P.O. (2003): Efficacy and safety of long-term bisphosphonates in postmenopauasal Osteoporosis. Expert. Opin. Phannacother., 4(12): 2253-2258.
21. Moallem, E., Silver, J., Kilav, R. and Naveh-Many, T. (1998): RNA protein-binding and post-transcriptional regulation of PTH gene expression by calcium and phosphate. J. Biol. Chem.; 273:5253-5259.
22. Ott SM. Bone histomorphometry in renal osteodystrophysemin Nephrol, 2009, 29: 122-32.
23. Pi-Sunyer, X.F. (1998): Clinical guidelines on the identification, evaluation and treatment of overweight and obesity in adults: the evidence report. Obes. Res., 6:152s-21 Os.
24. Raisz, L. (2005): Pathogenesis of Osteoporosis: Concepts, Conflicts and prospects. J.Clin. Invest., 115(2): 3318-3325.
25. Sambrook, P.N. (2000): Corticosteroid Osteoporosis: Practical implications of recent trials. J. Bone Miner. Res., 15:1645-1649.
26. Siek, G., Lawlor, J., Pelczar, D., Sane, M. and Musto, T. (2002): Direct serum total iron-binding capacity assay suitable for automated analyzers. Clinical chemistry 48:161-166.
27. Torres, P.U. and canaud, B.: How to slow down parathyroid hormone (PTH) secretion without the risk of inducing an adynamic bone disease. Nephrol.Ther.,(2005)l:s337-s341.
28. VanStaa, T.P., Leufkens, H.G. and Cooper, C. (2002): The epidemiology of corticosteroid induced Osteoporosis: A meta-analysis. Osteoporos. Int., 13:777-787.
29. Wick, M., Pingerra, W. and Lehman, N. (1996): Iron metabolism, diagnosis and therapy of anemia. Clin.Invest., 72: 511-515.
30. World Health Organization (WHO) (1994): Assessment of fracture risk and its application to screaning for postmenopausal Osteoporosis. Report of a WHO Study Group. World Health organization technical report series, 843:1-129.

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