Studying the impact of vitamin D deficiency in Iraqi acromegalic patients and its relation with some biochemical parameters

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Abstract:

Acromegaly is described as a less common chronic disease occursas a result ofover secretion of growth hormone (GH) often from a pituitary adenoma; it is connected to noticeable morbidity and increased mortality. Our study aimed to determine calcium (Ca) and vitamin D status inacromegalic patients regarding disease activity and estimating the relationship between vitamin D, IGF-1 and some biochemical parameters. Correlations of vitamin D with IGF-1, BMI, Ca, GOT, GPT, ALP, T3, T4, TSH, urea and creatinine were studied. The study groups comprise of 50 male, 25 acromegalic patients and 25 control male groups. The results of vitamin D measurements showed a decrease levels of vitamin D in all acromegalic patients and this revealed that patients with acromegaly have a high significant decrease ($p < 0.001$) of BMI, IGF-1, vitamin D, Ca, GOT and creatinine compared to control group, while significant increase for GPT, ALP, T3, T4 and urea for patients with acromegaly. However, the results showed a highly positive significant association between vitamin D and BMI, IGF-1, urea, creatinine and T4 levels, whereas demonstrated a highly significant positivecorrelation of vitamin D levels with GOT and ALP, also the results showed non-significant difference between vitamin D, Ca and T3 with a negative correlation with TSH also asignificant positive correlation between vitamin D and GPT appeared. We concluded that acromegalic patients were suffering from low vitamin D levels compared to the controls group and also suffering from lower kidney activity (hydroxylation at the position of 25-OH-D3 site) and this is evident from the high creatinine levels in the blood, which leads to the non-activation and even low levels of vitamin D and therefore acromemglic patients will suffer from vitamin D deficiency high risk thus the treatment procedures for acromegaly should include doses of active vitamin D, which reduces the risk of this deficiency.

Keywords: Acromegaly, Vitamin D,Calcium, Insulin-like growth factor-1

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Introduction:

Acromegaly is defined asa less common chronic disease caused by hyper-secretion of growth hormone (GH) usually from a pituitary adenoma and connected to noticeable morbidity and increased mortality [(1)]. Acromegaly affects nearly three cases in a million people each year in the western world, while the statistics estimate that the prevalence rates range between (5 to 9) cases out of every (10,000) of the total population in Iraq [(2)]. Insulin-like growth factor-1 (IGF-1) (mainlysynthesizedin peripheral tissues and liver) mostly mediates by effect of GH; however Anthony and Ioachimescu studied the relation between vitamin Dand (GH\IGF-1) axis [(3)]. The clinical diagnosis based on symptoms linked toGH excess, is usually delayed due to the insidious nature of the disease [(4,5)]. It was hypothesized thatIGF-1 and IGF binding protein-3 productionoccur in liver that stimulated by vitamin D through direct transcription regulation and\or by promoting GH stimulation. In kidney as well,IGF-1 increase stimulates the expression of 1α-hydroxylase which increases 1,25-dihydroxy vitamin D level that leads to alteration in calcium-phosphate balance in uncontrolled acromegaly [(7,8)].

Several studies illustrated that GH and IGF-1stimulate 25-dihydroxy vitamin D conversion to $1,25(OH)_2$vitamin D via calcium mediatedactivation of 1,α-hydroxylase in kidney which elevate calcium and phosphate absorption in the intestine [(8)]. Excess of 1,25-dihydroxy vitamin D in rare cases of hypercalcemia can also be encountered that used for acromegalypatients treatment. In acromegalyincreased bone desorptioncould have important role in calcium balance [(9,10)]. In acromegaly patients hypo-vitaminosis D reported [(11)]; also lower peripheral bio-availability of vitamin D had been showndue to the effect of GHexcess secretion on vitamin D binding protein [(12)].

Increased bone turnover had been related to active acromegaly disease [13] which causes secondary osteoporosis that lead to elevate fracture risk and even morbidity and mortality [14]. Cardiovascular and\or respiratory complications or neoplastic disease are common etiology of death in acromegalic patients. Many studies reported that increased development risk of benign and malignant tumors of various organs, particularly of the colon, thyroid gland, breast and prostate. IGF-1 elevated levels seem to be responsible for cancer risk. It is recalled that IGF-1 is amitogenic, anti-apoptotic and angiogenesis-promoting factor [15].

Many studies have demonstrated that hypercalcemia, hypercalciuria and meld hypophosphatemia in acromegaly are a result of 1,25-dihydroxyvitamin D3synthesis[6], further studies reported vitamin D deficiency in acromegalic patients[8]. An elevated risk of thyroid and colorectal cancer in patients with acromegaly had been shownby other studies[16]. The correlation between structural and endocrine changes in thyroid and patients demographic characteristics, GH levels, and IGF-1 levels in acromegaly remaininconsistent[17]. A web- based acromegaly patient registry has been developed from successive result of acromegaly to demonstrate the hypogonadism in male patients with microadenoms, especially. Also results revealed that hypersecretion of GH and prolactincontributes to the pathogenesis of acromegalyhypogonadisim and that could happen in macrodenoma patients even without hyperprolactinemia[18].

Materials and Methods

1- Subjects:

A group of patients attended the National Diabetes Center, Baghdad during the period from March to September 2018. A total of 50 males (aged 30-40 years) were enrolled in this study; 25 patients with acromegaly and 25 healthy male individuals as a control group. All patients were diagnosed by Physicians.

2- Biochemical Analysis:

Sera were separated from collected blood samples. Levels of GH, IGF-1, vitamin D, Ca, GOT, GPT, ALP, urea, creatinine TSH, T3 and T4 were determined. Serum IGF-1 was determined using IRAMA IGF-1 kit obtained from Beckman Coulter, using the RIA method. Calcium determined by derived O-cresol htaleinComplexone (CPC) method (Moorehead and Briggs, 1974) that allows total Ca concentration determination in serum. Serum GOT, GPT, ALP, urea and creatinine were determined using BioLABO kit from France. For GOT, GPT and ALP the method developed by Henry and Bergmeyer used (following modified IFCC recommendation). While for creatinine colorimetric reaction method was used (Jaffe reaction). Urea determined by enzymatic method. Vitamin D, T3, T4, TSH and were determined using VIDAS® kit obtained from Biomerux using EIFA method. BMI was determined by this equation:

$$\textbf{BMI= mass (kg)\ hight}^2\ \textbf{(m}^2\textbf{)}$$

3- Statistical Analysis:

Results are defined asMean±SEM, student t-test was applied to compare significance differences between all studied groups. *P*-value ($P<0.05$) and ($P<0.001$) considered statistically significant and highly significant, respectively.

Results:

Table (1) shows a high significant elevation in BMI in patients group G2 compared with control healthy group G1 ($P<0.001$), also a significantly higher IGF-1 concentration ($P<0.001$) appeared in patients with active acromegaly in contrast with control group as shown in Figure (1). Furthermore, Table (1) revealed that a highly significant decrease ($P<0.001$) in vitamin D level in patients group G2 compared with healthy group G1, while Calevel was significantly higher in patients group G2 compared to control group G1 ($P<0.001$) as illustrated in Figure (1) as well.

Table (1): Age, BMI and IGF-1 in control and acromegaly patientsexpressed as Mean±SEM

Parameter	G1 Control (Mean±SEM)	G2Patients (Mean±SEM)	*P*-value
Age	36.480 ± 0.753	37.120 ± 0.74	NS
BMI K/m^2	26.932 ±0.667	30.054± 0.664	HS
IGF-1(ng/ml)	304.08 ± 8.754	352.400± 14.918	HS
Vit D (ng/mL)	22.980 ± 0.777	13.692 ± 2.318	HS
Ca (ng/mL)	8.880 ± 0.128	10.220 ± 0.412	HS

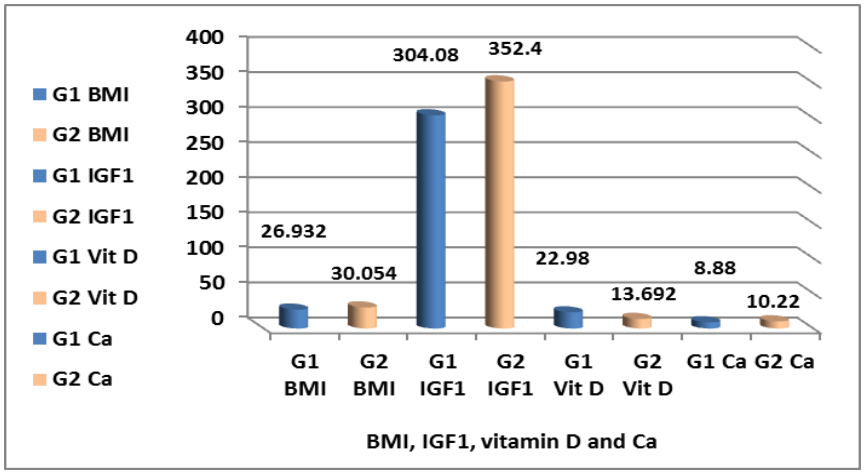

Figure (1):BMI, IGF-1, vitamin D and Ca levels in control G1 and patients G2 groups

A statistically significant differences (*P*>0.001) in GOT, GPT, ALP, urea creatinine and hormones of thyroid had been shown in acromegaly patients as compared to control as listed in Table (2). On the other hand, mean levelsof GOT and creatinine werelowersignificantly (*P*>0.001) in control group compared to patients with acromegaly as shown in Figures (2, 3 and 4).

Table (2): Values of GOT, GPT, ALP, Urea, Creatinine, TSH, T3 and T4 in control and patients with acromegaly expressed as Mean±SEM

Parameter	G1 Control (Mean ± SEM)	G2Patients (Mean ± SEM)	*P* - value
GOT U\L	23.176 ± 1.103	28.840 ± 1.308	HS
GPT U\L	28.352 ± 1.092	29.292 ± 1.949	NS
ALP U\L	188.00 ± 14.096	234.00 ± 27.278	NS
Urea (mg/dl)	25.612 ± 0.738	39.888 ± 2.744	NS
Creatinine (mg/dl)	0.7356 ± 0.032	1.00 ± 0.09	HS
TSH Pmol\L	2.648 ± 0.202	9.570± 2.795	S
T3 Pmol\L	5.436 ± 0.202	5.116 ± 0.444	NS
T4 Umol\L	16.152 ± 0.515	16.344 ± 1.652	NS

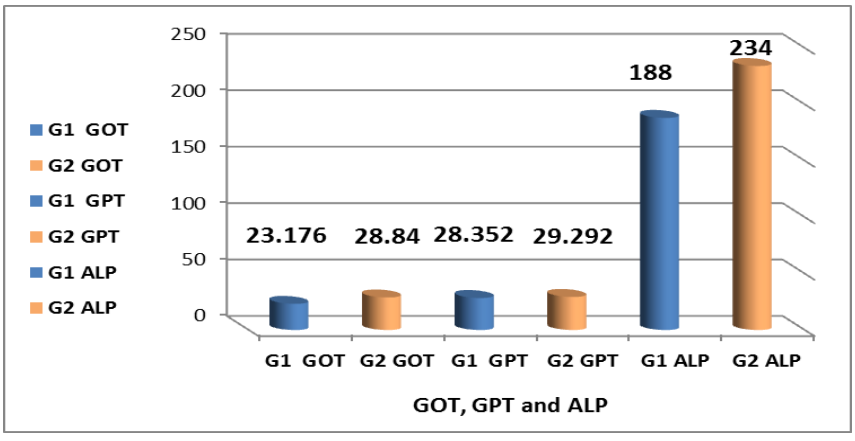

Figure (2): Levels of GOT, GPT and ALP in control G1 and patients G2 groups

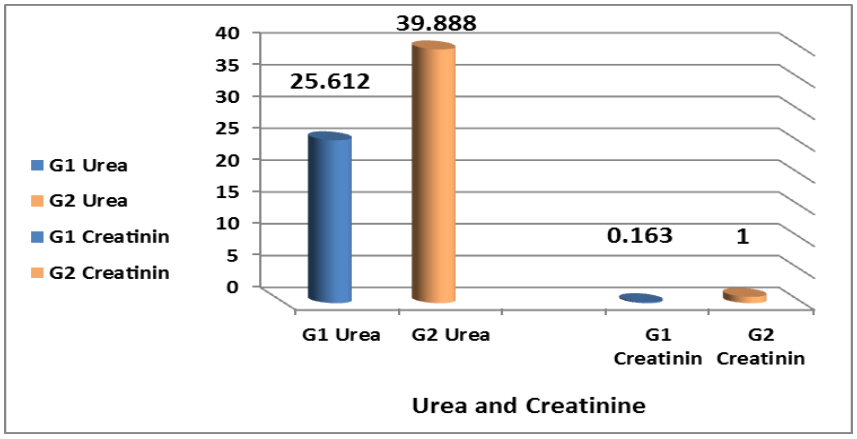

Figure (3): Levels of Urea and creatinine in control G1 and patients G2 groups

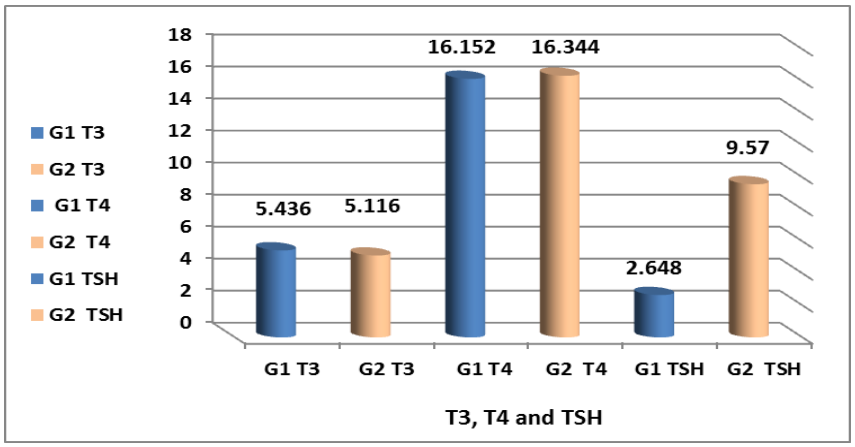

Figure (4): Levels of T3, T4 and TSH in control G1 and patients G2 groups

As presented in the table (3), a high positive significant correlation is shown between vitamin D and BMI, IGF-1, urea, creatinine and T4 levels, while asignificant highly negative correlation found for levels of vitamin D with GOT and ALP, also the results show a positivenon-significantcorrelation between vitamin D and Ca and T3, negative correlation with TSH and significantly negative correlation with GPT, Figures (5, 6, 7 and 8).

Table (3): Pearson correlation and *P*-value for vitamin D with the studied parameters

Parameter	Pearson Correlation	*P*- value
BMI	0.07	HS
IGF-1	0.563	HS
Ca	0.572	NS
GOT	- 0.063	HS
GPT	- 0.386	S
ALP	- 0.148	HS
Urea	0.688	HS
Creatinine	0.707	HS
TSH	- 0.278	NS

T3	0.442	NS
T4	0.324	HS

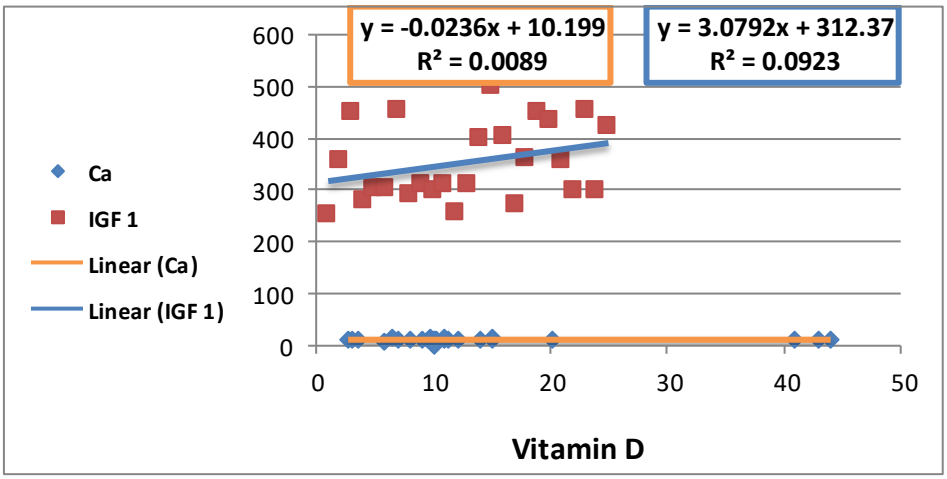

Figure (5): Correlation relation between vitamin D and calcium level

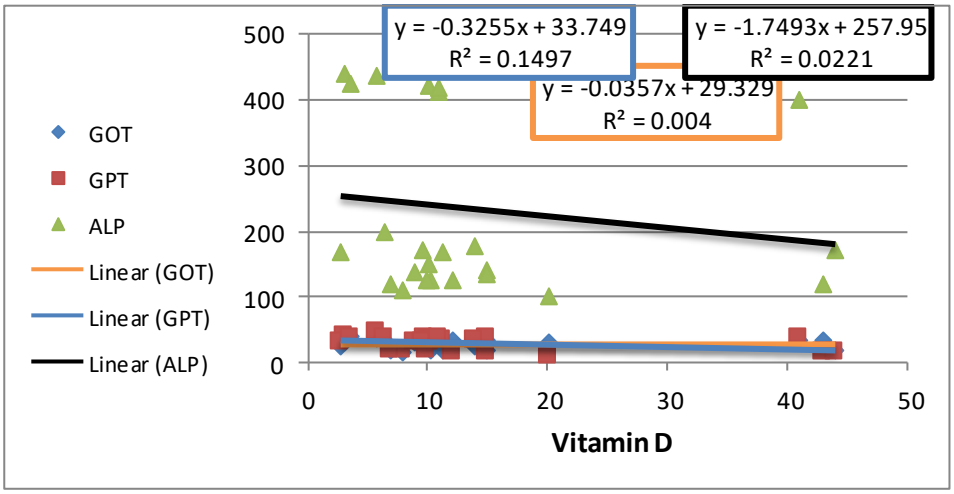

Figure (6): Correlation relation between vitamin D and levels of GOT, GPT and ALP

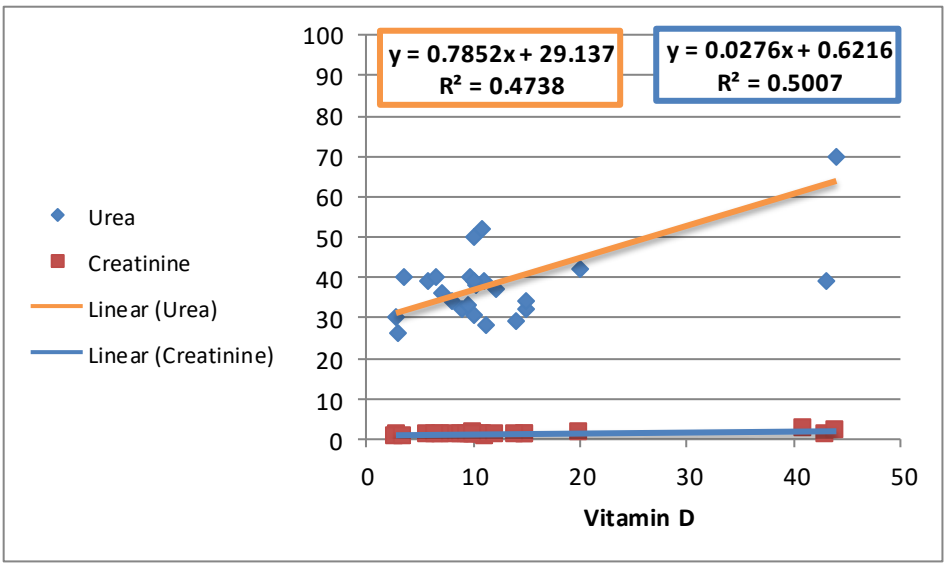

Figure (7): Correlation relation between vitamin D and levels of urea and creatinine

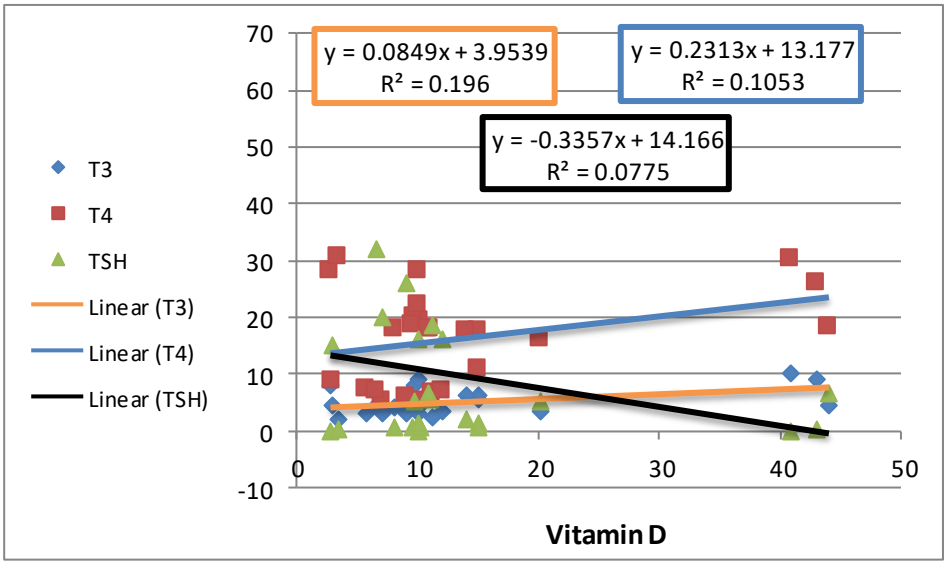

Figure (8): Correlation relation between vitamin D and levels of T3, T4 and TSH

Discussion:

Vitamin D in Acromegaly Patients:

Various medical conditions are associated with the deficiency of Vitamin D like musculoskeletal disorders, metabolic diseases, infection and cardiovascular disease. The decrease in serum vitamin D levels in acromegaly patients could be attributed to inadequate exposure to sunlight and nutritional deficiency, or to etiopathogenetic reasons. Also it may be concluded that insufficiency of vitamin D could cause fatigue, increased disease activity and decreased quality and functional capacity of life.

The optimal indicator of vitamin D status is 25-hydroxyvitamin D.Vitamin D3production occurs in the skin through a two-step process. A non-enzymatic stepduring exposure to ultraviolet rays in sunlight to convert 7-dehydrocholesterol to pre-vitamin D3 and a temperature-dependent reaction forming vitamin D3 from pre-vitamin D3.

Vitamin D3bound to vitamin D-binding protein (DBP)is then transported to the liver and metabolized to 25(OH)D, via enzymatic process. Alternatively, vitamin D (D2 or D3), can be absorbed in intestine. By the action of 25-hydroxylaseeither D2 or D3 is metabolizedto 25-hydroxy vitamin D2 or hydroxyl vitamin D3, respectively. 25-Hydroxy vitamin D2 and D3 bound to DBP enter the circulation.25(OH)Dwithin the renal tubular cell is released from the binding protein, following up take of the DBP-25(OH)D complex by megalin/cubilin in renal tubules and the parathyroid glands [19].

Endocytic receptors are responsible for the intracellular formation of biologically active 1-α,25-dihydroxyvitamin D3from 25-hydroxyvitamin D3. The dysfunction of these receptors found in patients with diabetic nephropathy, may explain vitamin D deficiency that occurs in these patients.

After catabolism, 1-α-hydroxylase and 24- α-hydroxylase act on 25(OH) D to generate 1, 25 dihydroxy vitamin D and 24, 25 dihydroxy vitamin D [20].Several chronic conditions like nephrotic syndrome, critical illness and end stage liver diseasewhere levels of parathyroid hormone are not raised are recognized by low 25(OH) D levels [21].Moreover, vitamin D may affect metabolic disease, diabetes, metabolic syndromeand insulin resistance [22]. In our study, concentration of vitamin D was decreased that is in agreement with other studies [12, 23] which found that vitamin D deficiencyor gonadal status is not related to the differences in gene expressions in patients with acromegaly.

Association of Vitamin D with IGF-1:

Serum levels of IGF-1 were highly significant increase in acromegaly patients compared with control group (P<0.001). According to related studies[7], it can be assumed thatliver production of IGF-I and IGFBP-3 ispromoted by vitamin D via inducing relevant genestranscription directly and/or by enhancing GH stimulation.

Acromegaly could be caused by GH excessive excretion and IGF-1 levels elevation.

An exacerbate complications occurs if diagnosis delayed due to GH hypersecretion. Thus, timing is crucial in acromegaly clinical and biochemical diagnosis. Correlation between acromegaly-associated factors and serum IGF-1has been noted, so serum IGF-1 levels should be measured when acromegaly is suspected[24]. In acromegaly diagnosis,IGF-1 level is an ideal screening test rather than random GH levels because with low IGF-1 levels acromegaly is extremely rare[25].

Vitamin D and BMI:

Serum 25(OH)D3 affected by BMI could be explained by high contents of body fat that storeslipid-soluble vitamin D.In animal models, about 10-12% of vitamin D dose supplemented can be accumulated in adipose tissue, where vitamin D release is extremely slow from fat and related to vitamin concentration in the adipose tissue[26]. This process may protect the body from vitamin D active forms toxicity effects and maintain optimal level in the blood; while extra body fat results in its low availability and elevated restoration and, as a result, low serum 25(OH)D levels [12, 26].

Vitamin D and Calcium:

Acromegalic patients may have hypercalcemia, Shah et al.[27] reported two diagnosed acromegaly cases have mild hypercalcemiawith normal serum 1, 25(OH)D2 levelsand total 25(OH)D. In the kidney, IGF-1may have appositive influence on1, 25(OH)D2synthesis by stimulating 1-α hydroxylase activity [7]. Same elevated levels 25(OH)D and1, 25(OH)D2 in active and inactive acromegaly patients with no hypercalcemiaobserved[9].

Our study agrees withother studies which revealed that total concentrations of 25(OH)D depend on alterations in VDBP levels[6, 9, 23].

Association of Vitamin D with Liver Function (GOT, GPT and ALP):

Target tissue for vitamin D production is liver that isconsidered essential source of circulating IGF-1 [7]. Serum GOT enzyme can associate with red blood cells, skeletal muscles action, heart muscles, brain and kidney; while GPT enzyme is associates with diseases incidence like hepatitis and cirrhosis. One of the previous studies suggested that GOT and GPT can also be associated in acromegaly occurrence [28]. Our study agrees with previous studies in significant elevated GOT, GPT and ALP activity levels in acromegalic patients. Another study showed that pituitary surgery in patients with acromegaly led to significant decrease in GPT levels [29].

Association of Vitamin D with T3, T4 and TSH:

Our studyrevealed significant increase levels of T4 and TSH in patients group G2 in comparison with control group G1, while T3 levels decreased; these results doesn't agree with those reported by Muscogiuri[30] and Carnevale[31] that didn't discover any correlation between vitamin D and PTH. However, many studies

showed that acromegalic patients are atincreased incidence of thyroid cancer and thyroid nodular disease[32], previous studies have shown increase [33], decrease[10] or no change [34] in PTHlevelsafter treatment of acromegaly in consistencies that may reflect the single time point sampling methodology used in the study.

Association of Kidney Function (Urea and Creatinine) with Vitamin D:

This study shows non-significant increase in urea levels ($P>0.05$) among acromegaly patients group in comparison with control group, and highly significant increase in creatinine levels ($P<0.001$). Impairment of any functions of kidney is usually indicated with high value of creatinine; while blood urea nitrogen (BUN) alteration indicates chronic and acute renal disease,so our result doesn't agree with Kamenicky[9]that found concentration of creatinine was lower before patient's acromegaly treatment.

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