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RELATIONSHIP BETWEEN VITAMIN D DEFICIENCY AND PHYSIOLOGICAL BLOOD PARAMETERS IN HYPOTHYROIDISM PATIENTS

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ABSTRACT : Vitamin D is "a fat-soluble steroid hormone" ingested in the diet but the major produced occur through the skin after exposure to the solar ultraviolet rays in sunlight. Vitamin D deficiency "VDD" is now commonly accepted has been associated with a number of clinical and endocrine disorders. This study suggested a high degree association between Vitamin D deficiency "VDD" and risk of anemia in individuals with hypothyroidism. In the present study, after blood sample was collected from twenty adults (age 18 - 45 years) was conducted on newly clinical and laboratory diagnosed patients of hypothyroidism and twenty healthy control group. Thyroid hormone analysis to determine the hypothyroidism and estimate of serum 25(OH)D3 levels with blood measurement including hemoglobin concentration, ferritin, MCH, MCV, HCT also were determined. The results were revealed Vitamin D was significantly decreased (9.629 ± 2.773ng/ml) in patients than control (38.15 ± 22.73ng/ml) at (P < 0.05) and serum TSH levels were significantly higher in cases (21.98 ± 17.24µIU /ml) as compared to healthy group (2.059 ± 2.334µIU/ml) at (P < 0.05). And showed the results of the current study significant effects on hormonal and physiological blood criteria (HB, ferritin, MCH, MCV, HCT) when compared with one another and with the healthy controls. The study concluded that the relationship between Vitamin D deficiency and blood physiological parameters in that patients of hypothyroidism. These findings could have been useful in the diagnosis pathogenesis of hypothyroidism and anemia and the use in the supplementation of Vitamin D supplements as therapy for patients with anemia and hypothyroidism.

Key words : Vitamin D deficiency, blood parameters, Hypothyroidism, Hemoglobin.

INTRODUCTION

Vitamin D is ''a fat-soluble steroid hormone" ingested in the diet but the major produced occurthrough the skin after exposure to the solar ultraviolet rays in sunlight (McCarty *et al*, 2013). Vitamin D is converted by the liver to inactive form ''25(OH) Vitamin D, it has a half-life of 15 days" and is metabolized in kidneys to the active form "1,25-dihydroxyvitamin D" by the enzyme "25-hydroxyvitamin D-1α-hydroxylase (CYP27B1)" (Lips, 2006). Only serum "25(OH) Vitamin D3 also called (calcidiol)" considered as the real indicator of the total Vitamin D stores and is used for clinical evaluation of the Vitamin D3 status in body, while circulating 1,25(OH)D is not a good indicator of Vitamin D status because ofithashalf-life of 15 hours shorter than "25(OH) Vitamin D'' (IMFNB, 2010). The normal range of Vitamin D assay has approximately 20-60ng/dL, this range may be too low for many serum patients, so when the concentration is less than 30ng/mmol leading to the case called "Vitamin D deficiency VDD" (Lee *et al*, 2008) and if it is less than 10ng/ml (12.5nmol/L) signifies severe deficiency (William, 2004). The effects ofVitamin D mediates have done by binding to "vitamin D receptor (VDR)" and then the activation of "VDR-responsive genes" (Theodore; William and Stephen). It is only the ''1, 25-OH Vitamin D'', which is biologically active andit acts to allow for absorption of calcium (Ca^{++}) from the intestinal tract ''the principled role ofVitamin D is to be regulating calcium homeostasis''. Therefore, patients with low Vitamin D levels in the blood will have low calcium levels in severe cases ''get rickets in children" or "osteomalacia in adults'' that occurs when the bone bows out and is poorly formed, while in the mild cases of Vitamin D deficiency (VDD), ''osteoporosis" occurs and in the chronic conditions ''cancer and metabolic syndrome", in addition to the Vitamin D deficiency may occur in patients with malabsorption from their intestine, such as in the case "the autoimmune disease called Celiac Disease", also it occurs frequently in patients with thyroid problems (Holick *et al*, 2006; Dusso *et al*, 1994). As well as to the main role for Vitamin D3 in regulation of bone and mineral metabolism, it also has an important role in regulating

cellular proliferation and differentiation (Robert *et al*, 1980), because of the steroid hormone receptors (VDR) are present in different body cell tissueslike myocardium, pancreas, reproductive system and thyroid gland etc (Norman, 2006). So, the decreasing inconcentration of Vitamin D will be leading to exacerbate the systemic abnormalities associated with hypothyroidism (Wang *et al*, 2008; Chopra *et al*, 2011). Hypothyroidism is defined as "a deficiency of thyroid activity through reducing the secretion of both T4 and T3 thyroid hormone concentrations leading to hyper secretion of pituitary TSH by negative feedback mechanism casing increase in serum TSH levels. One of the most important effects of Vitamin D that is in the proliferation and differentiation of cells of the bone marrow casing anemia, so there is a significant relationship between Vitamin Ddeficiency and anemia (Sim *et al*, 2010) was recorded in the world, as the role of Vitamin D in "erythropoiesis" (Saab *et al*, 2007) and calcitriol is involved in ''haematopoiesis" then affects marrow function (Norman, 2006). About 50% of all anemia cases are diagnosed as ''iron deficiency anemia (IDA)", patients with Iron deficiency anemia (IDA) their hemoglobin less than (<12 mg/dl) (Zimmermann *et al*, 2007), so an increasing body of evidence indicates that VDD was associated with increased risk for anemia. Generally, the aim of study was established by estimate serum Vitamin D_3 concentrations, the relationship between Vitamin D deficiency andblood physiological parameters in newly diagnosed hypothyroidism patients on the basis of raised thyroid stimulating hormone levels compared with healthy controls.

MATERIALS AND METHODS

In the present study, after blood sample was collected from twenty adults (age range between 18 and 45 years) was conducted on newly clinical and laboratory diagnosed patients of hypothyroidism and twenty healthy control group. The diagnosis was based on detailed history of any chronic diseases such as "diabetes mellitus, hypertension, liver disease, renal disease, metabolic bone disorders, malignancy, hypo- or hyperparathyroidism, vitamin and mineral deficiency, steroid and anti-osteoporotic therapy'' were excluded from the study. Thyroid hormone analysis to determine the hypothyroidism and estimate of serum 25(OH)D_3 levels with blood measurement including hemoglobin concentration, ferritin, MCH, MCV, HCT also were determined. Serum was separated and stored at -20°C for estimation of 25(OH)D_3 levels, blood parameters and thyroid hormones. Serum 25(OH)D_3 level was measured by ELISA assay kit, which was designed for the determination of 25(OH)D_3 in human samples (Eagle Biosciences Inc., MA, USA), ferritin (FER) level was determined in human serum by VIDAS FER kit (distributed by bioMerieux SA 376 Chemin de ĐÈOrme 69280 Marcy-ĐÈEtoile - France) using the ELFA technique (Enzyme Linked Fluorescent Assay), thyroid stimulating hormone (TSH) level was measured by using the VIDAS® TSH assay, which is intended for use on the instruments of the VIDAS family (Vitek®Immuno Diagnostic Assay system) as "an automated quantities enzyme-linked fluorescent immunoassay (ELFA) for the determination of human thyroid stimulating hormone (TSH) concentration in human serum that is intended for use as an aid in the diagnosis of thyroid disorders" (distributed by bioMerieux, Inc. 100 Rodolphe Street Durham, North Carolina 27712-USA). Blood parameters were measured in complete blood count patients and healthy controls group by using Auto Hematology Analyzer (Diagon® Ltd D-Cell 60).

Statistical analysis

After maintain the studied data on excel spread sheet, the results were analyzed statistically using Graph Pad prism 5 program appoint the arithmetic mean value and standard errors (SE) and standard deviation (SD) of all variables and test the significance treatment groups used one-way ANOVA analysis of variance Tukeyto compare the hematology and biochemical parameters between patients and healthy controls. An association between study variables was assessed using MegaStat's correlation analysis. Differences were considered statistically significant at $p<0.05$.

RESULTS

The circulation of biochemical characteristics and complete blood count parameters in patients and healthy control groups are shown in Table 1, revealed that serum Vitamin D was significantly decreased (9.629 ± 2.773ng/ml) in patients than control (38.15 ± 22.73ng/ml) at ($P < 0.05$). Additionally, Table 1 shows the results obtained for serum TSH levels were significantly higher in cases (21.98 ± 17.24µIU /ml) as compared to healthy group (2.059 ± 2.334µIU/ml) at ($P < 0.05$), so the mean levels of serum vitamin D in hypothyroid patient group was significantly decreased (9.629 ± 2.773ng/ml) compared to healthy adults (38.4±22.73 ng/ml).

On MegaStat's correlation analysis depicted a significant and negative correlation between levels of Vitamin D and TSH ($r = 0.0485$, $p< 0.05$) (Fig. 1), also a negative correlation between levels of TSH withHb ($r = 0.0153$, $p<0.05$) and Ferritin ($r = 0.0760$, $p<0.05$) respectively (Figs. 4, 5), while the obtained data indicated that Vitamin D has positive correlation with Hb (r =

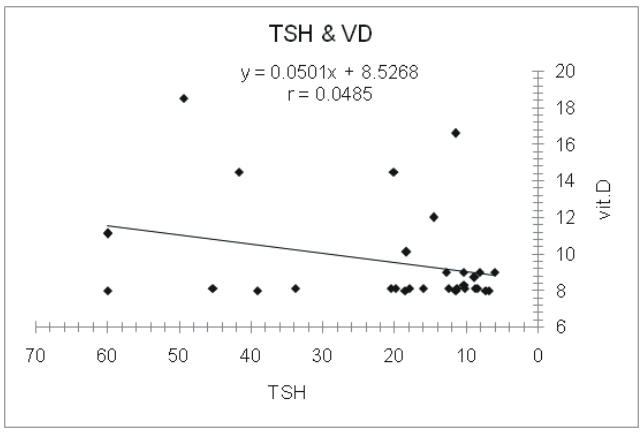

Fig. 1 : Scatter diagram showing correlation between Vit D and TSH (r = 0.0485, p< 0.05).

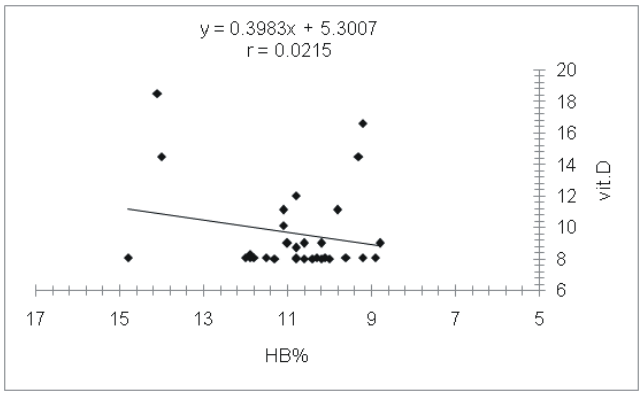

Fig. 2 : Scatter diagram showing correlation between Vit D and Hb% (r = 0,0215, p<0.05).

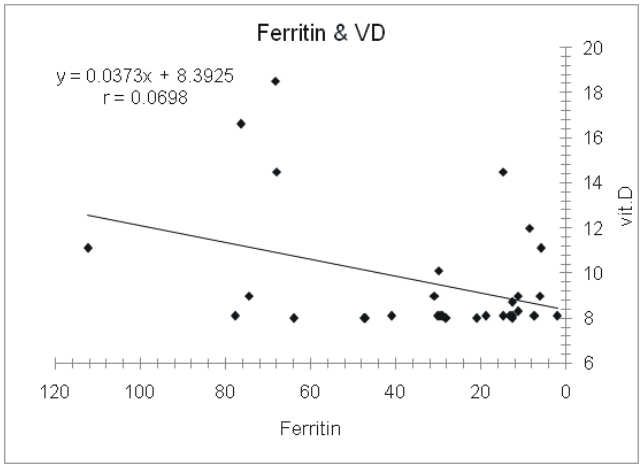

Fig. 3 : Scatter diagram showing correlation between Vit D and Ferritin (r = 0.0698, p< 0.05).

0,0215, p<0.05) and Ferritin level (r = 0.0698, p< 0.05) in patients respectively (Figs. 2, 3). Regarding to the results in Fig. 6 was shown a significant at P < 0.05 in positive correlation with Hb and ferritin level (r = 0.0551, p<0.05) in patient groups.

DISCUSSION

Vitamin D deficiency ‘‘VDD” is now commonly

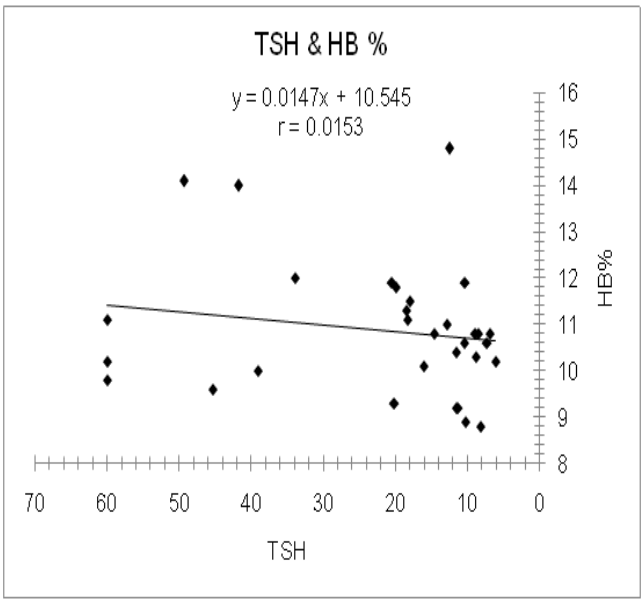

Fig. 4 : Scatter diagram showing correlation between TSH and HB% (r = 0.0153, p<0.05).

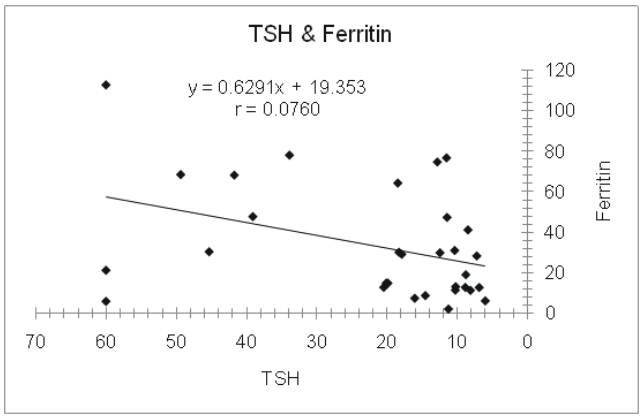

Fig. 5 : Scatter diagram showing correlation between Ferritin and TSH (r = 0.0760, p<0.05).

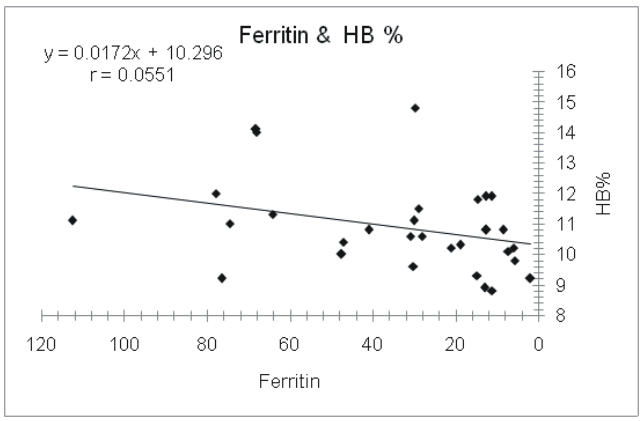

Fig. 6 : Scatter diagram showing correlation between Ferritin and HB% (r = 0.0551, p<0.05).

accepted has been associated with a number of clinical and endocrine disorders. This study suggested a high degree association between Vitamin D deficiency “VDD” and risk of anemia in individuals with hypothyroidism,

Table 1 : Mean ± SD, ± SE values of all parameters in studies patients compared with healthy control groups.

Parameter	Patients	Healthy groups
Vitamin D (ng/ml)	9.629 ± 2.773± 0.4981	38.15 ± 22.73± 5.083
TSH (ìÉU/ml)	21.98 ± 17.24± 3.096	2.059 ± 2.334± 0.522
Ferritin(ng/ml)	33.18 ± 27.81± 4.995	84.78 ± 120.5± 26.95
Hb %(g/dl)	10.87 ± 1.444± 0.2594	12.73 ± 1.471± 0.3289
MCV(û)	82.37 ± 6.496± 1.167	86.70± 1.435± 0.3208
MCH(pg)	25.63 ± 2.563± 0.460	26.65 ± 0.8488± 0.1898
HCT(pg)	37.56 ± 4.922± 0.8841	40.87 ± 2.513± 0.5619

Vitamin D: 25-hydroxyvitamin D3, **TSH :** thyroid stimulating hormone, **Hb:** Hemoglobin, **MCV:** mean corpuscular volume, **MCH:** mean corpuscular hemoglobin.

so that is observed in the current study low levels of serum 25(OH). Vitamin D has often been a significant correlation ($p < 0.05$) with deficiency of serum ferritin and hemoglobin Hb levels and increased serum TSH levels compared with those normal serum 25(OH) vitamin D levels. This is agreed with other studies that were showed an association between Vitamin D3 level and iron deficiency anemia patients ''without and chronic kidney disease" (Peristgein *et al*, 2011; Patel *et al*, 2010), while Sonawane *et al* (2017) suggested that a significant relationship between deficiency of vitamin D and increasing serum TSH levels (Sonawane *et al*, 2017). On the different causes leading to the vitamin D deficiency like reduce sunlight exposure and poor intestinal absorption of Vitamin D (Theodore) that the main action of vitamin D is the regulation of calcium and bone marrow metabolism "including cellular proliferation and differentiation" (Arabi *et al*, 2010; Norman, 2008). Importantly, its receptors ''VDR", which is found in must cell types including thyroid gland [21]and bone marrow (Kiss *et al*, 2011), the levels of 1, 25 (OH) vitamin D (active form of Vitamin D) are several hundred folds higher in bone marrow compared to plasma so it called "steroid hormone receptors similar to thyroid hormone receptors" because of vitamin D made up from cholesterol of the body skin helping by sunlight then casing stimulate "erythroid precursors''. In order to function, results of the present study reported a negative correlation between serum Vitamin D_3 levels and TSH in hypothyroid patients by meaning "low levels of Vitamin D increase the risk of hypothyroidism". This may be occur by interacting with its receptors in the thyroid gland so it has active role in maintaining a euthyroid state (Zaletel and Gaber, 2011). Therefore, the deficiency of vitamin D VDD may be leading to decrease of thyroid hormone levels including T3 and T4 with increasing of TSH levels. As well as other studies were showed that the patients of "autoimmune thyroid disease having low levels of Vit D3 (Unal *et al*, 2014). Other studies like an experimental study by Byron Richards (2008) studied, that was showed a lack of Vit D leading to the possibility of decrease thyroid hormones (Byron, 2008), so the significant ($p < 0.05$) negative correlation between vitamin D and TSH indicates the correlation between hypothyroidism and Vitamin D. On the other hand, the data obtained that revealed decreasing in the blood parameters including HB hemoglobin, ferritin, MCH, MCV and HCT correlated with vitamin D deficiency ''VDD", this study same as the results of recent studies that have shown a strong relationship between Vitamin D deficiency and iron deficiency anemia in female population of Kerbala city in Iraqi patients that were having significantly low levels of its serum vitamin D_3 than that in healthy (Norman, 2006) and also agreed with the results of an experimental studied John Sim *et al* (2010), which demonstrated an association between vitamin D deficiency and a higher risk of anemia with lower hemoglobin levels (Sharara *et al*, 2017). However, the relationship between VDD and anemia may be resulting from the effects vitamin D on bone marrow metabolism like a direct effect on erythropoiesis processes "the mechanism of RBCs formation" and then its effects on hemoglobin levels so it has significantly and positive correlation ($p< 0.05$) between Vitamin D and Hb, ferritin (Iron), WBCs and other blood parameters. Additionally, other studies were found a significant association of Iron deficiencyanemia and low Vitamin D levels among1-12 months aged infants "due to its reduced intestinal absorption of Vit D caused by Iron deficiency" (John *et al*, 2010). Hence, the intestinal absorption of vitamin D may be impaired due to Iron deficiency as same as the absorption fat and vitamin A (Heldenberg *et al*, 1992). Moreover, the deficiency of vitamin D has a strong effect in the reduce proliferation and activation of RBCs, its severe affects in the decrease synthesis and metabolism of Iron and hemoglobin ultimately reducing bone mineral density, thus affecting "bone formation and bone reabsorption" (Katsumata *et al*, 2009). On the other hand, there is a significant positive correlation of TSH levels with ferritin and Hb leading to the thinking that a relationship between anemia and thyroid dysfunction that

has been demonstrated by the study (Nawras, 2017), which was shown increasing of levels of TSH in female patients with hypothyroidism beside reduced of levels of HB as compared to healthy controls (Nawras, 2017), thus also agreed with Beard *et al* (1990), which was found in the study that T3 and T4 levels were significantly decrease, TSH levels were significantly higher in anemic women, and they have determined that T3 levels have been increased by treatment with iron (Beard *et al*, 1990; Lippi *et al*, 2008).

CONCLUSION

We can conclude from the current study that was relationship between vitamin D deficiency and blood physiological parameters in that patients of hypothyroidism. These findings could have been useful in the diagnosis pathogenesis of hypothyroidism and anemia and in the supplementation of vitamin D supplements as therapy for patients with anemia and hypothyroidism.

REFERENCES

Arabi A, El Rassi R and Fuleihan G E (2010) Hypovitaminosis D in developing countries—prevalence, risk factors and outcomes. *Nature Reviews Endocrinology* **6**(10), 550-561.

Beard J L, Tobin B W and Smith S M (1990) Effects of iron repletion and correction of anemia on norepinephrine turnover and thyroid metabolism in iron deficiency. *Proc. Soc. Exp. Biol. Med*. **193**, 306-312.

Byron Richards (2008) Low Vitamin D contributes to Thyroid Problems. *Health News* 2008.

Chopra S, Cherian D and Jacob J J (2011) The thyroid hormone, parathyroid hormone, and vitamin D associated hypertension. *Indian J. Endocrinol. Metab.* **15**(14), S354-S360.

Dusso A, Brown A and Slatopolsky E (1994) Extrarenal production of calcitriol. In *Seminars in nephrology.* **14**(2), 144-155.

Heldenberg D, Tenenbaum G and Weisman Y (1992) Effect of iron on serum 25-hydroxy vitamin D and 24, 25-dihydroxy vitamin D concentrations. *Am. J. Clin. Nutr.* **56**, 533–536.

Holick Michael F and Michele Garabedian (2006) Vitamin D: photobiology, metabolism, mechanism of action,and clinical applications. Primer on the metabolic bone diseases and disorders of mineral metabolism. 6th ed. Washington, DC: *American Society for Bone and Mineral Research* **2006**, 106-14.

Institute of Medicine, Food and Nutrition Board (2010) Dietary Reference Intakes for Calcium and Vitamin D. Washington, DC: national Academy Press.

Kiss Z, Ambrus C, Almasi C, Berta K, Deak G, Horonyi P and Mucsi I (2011) Serum 25(OH)-cholecalciferol concentration is associated with hemoglobin level and erythropoietin resistance in patients on maintenance hemodialysis. *Nephron Clin. Pract*. **117**, c373–c378.

John J Sim, Peter T Lac, In Lu A Liu, Samuel O Meguerditchian, Victoria A Kumar, Dean A Kujubu and Scott A Rasgon (2010) *Vitamin D deficiency and anemia : a cross-sectional study* **89**, 447-452.

Katsumata S, Katsumata-Tsuboi R, Uehara M and Suzuki K (2009) Severe iron deficiency decreases both bone formation and bone resorption in rats. *J. Nutr.* **139**, 238–243.

Lippi G, Montagnana M, Salvagno G L and Guidi G C (2008) Should women with abnormal serum thyroid stimulating hormone undergo screening for anemia? *Arch. Pathol. Lab. Med*. **132**, 321-322.

Lips P (2006) Vitamin D deficiency and physiology. *Progress in Biophysics and Molecular Biology* **92**(1), 4-8.

Lee J H, O'Keefe J H, Bell D, Hensrud D D and Holick M F (2008) Vitamin D deficiency: an important, common, andeasily treatable cardiovascular risk factor?. *Journal of the American College of Cardiology* **52**(24), 1949-1956.

McCarty D E, Reddy A, Keigley Q, Kim PY, Cohen S and Marino A A (2013) Nonspecific pain is a marker for hypovitaminosis D in patients undergoing evaluation for sleep disorders : a pilot study. *Nature and science of sleep* **5**, 37.

Norman A W (2008) From vitamin D to hormone D: fundamentals of the vitamin D endocrine system essential for good health. *The American Journal of Clinical Nutrition* **88**(2), 491S-499S.

Norman A W (2006) Vitamin D receptor : new assignment for an already busy receptor. *Endocrinology* **147**(12), 5542- 5548.

Nawras N Bashboosh (2017) Correlation between Hypothyroidism and Iron deficiency anemia in female patients. **6**(7), 80-89.

Perlstein T S, Pande R, Berliner N and Vanasse G J (2011) Prevalence of 25-hydroxy vitamin D deficiency in subgroups of elderly persons with anemia : association with anemia of inflammation. *Blood* **117**(10),. 2800-2806.

Patel N M, Gutiérrez O M, Andress D L, Coyne D W, Levin A and Wolf M (2010) Vitamin D deficiency and anemia in early chronic kidney disease. *Kidney International* **77**(8), 715-720.

Robert K Murray, David A Bender, Kathleen M Botham, Peter J Kennely, Victor W Rodwell and Anthony Weil P (1980) Micronutrients : Vitamins and Minerals- Harper's Illustrated Biochemistry. **29**, 529-531.

Sim J J, Lac P T, Liu I L, Meguerditchian S O, Kumar V A, Kujubu D A and Rasgon S A (2010) Vitamin D deficiency and anemia: a cross-sectional study. *Ann. Hematol.* **89**, 447-452.

Saab G, Young D O, Gincherman Y, Giles K, Norwood K and Coyne D W (2007) Prevalence of vitamin D deficiencyand the safety and effectiveness of monthly ergocalciferol in hemodialysis patients. *Nephron Clinical Practice* **105**(3), c132-8.

Sonawane S, Bora B, Shrikhande D Y, Bansal Sahil and Kumar Prabhat (2017) Vitamin D Deficiency and its association with thyroid diseases. *International journal of Contemporary Medical Research* **4**(8).

Sharara F A, Fadhil J Al-Tu'ma1 and Lamyaa Abd Alkarem (2017) Deficiency of Vitamin D and Iron in Anemic female Iraqi patients. *International Journal of Pharmacy & Pharmaceutical Research*, **8**(3), 87-96.

Theodore C Friedman (.....) *Vitamin D Deficiency and Thyroid Disease*. www.goodhormonehealth.com/ Vitamin D.

Unal A D, Tarcin O, Parildar H, Cigerli O, Eroglu H and Demirag N G (2014) Vitamin D deficiency is related to thyroid antibodies in autoimmune thyroiditis. *Centr. Eur. J. Immunol*. **39**(4), 493-497.

Wang T J, Pencina M J, Booth S L, Jacques P F, Ingelsson E and Lanier K (2008) Vitamin D Deficiency and risk of cardiovascular disease. *Circulation* **117**, 503-511.

William J Marshall and Stephen K Bangert (2004) The clinical biochemistry of nutrition, *Clinical Biochemistry – Metabolic and clinical aspects*. 2nd Edition.

William J Marshall and Stephen K Bangert (....) Calcium, Magnesium and Phosphate. *Clinical Biochemistry – Metabolic and clinical aspects*. **2**, 99-100.

Zaletel K and Gaber S (2011) Hashimoto's Thyroiditis : from genes to the disease. *Current Genomics* **12**, 576-588.

Zimmermann M B and Hurrell R F (2007) Nutritional iron deficiency. *The Lancet*. **370**(9586), 511-20.