

Study of Vitamin D Status in Normal Healthy Individuals and Its Association with Parathyroid Hormone Levels

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Abstract: Vitamin D is a prohormone known to play a key role in phosphocalcic metabolism. The aim of the study is to determine the levels of vitamin D and to correlate the values of 25-hydroxy vitamin D levels with the levels of parathyroid hormone, Calcium (Total and Ionized) and Phosphorus. The study was conducted in the normal healthy individuals of Chennai population. 25-OH vitamin D and intact parathyroid hormones were analyzed by chemiluminescence method with ADVIA CENTAUR XP system. Among the study population about 21% of individuals are vitamin D deficient (group- 1) and 43% individuals are vitamin D insufficient (group-2) and 36% of the population are vitamin D sufficient (group-3) . A significant inverse relation was observed between vitamin D and intact parathyroid hormone levels in all the three groups is found to be statistically significant with p value of 0.002. In group 1 the total calcium level is decreased which is statistically significant with p value <0.001. The ionized calcium and phosphorus are found to be in normal range in all the three groups and it is also statistically insignificant with the p value 0.887 and 0.337 respectively. In this study the vitamin D levels have negative correlation with intact parathormone levels. As vitamin D level decreases there is a proportionate increase in parathormone level.

Keywords: Vitamin D, PTH, 25-(OH) D, Calcium, Vitamin D deficiency.

INTRODUCTION

Vitamin D stands at the frontline of current scientific endeavors being a topic of interest to the medical researchers all over the globe. D vitamin D deficiency is a risk factor in humans, right from conception and throughout lifespan.

Globally about one billion people are known to have vitamin D insufficiency or deficiency [1]. Prevalence of vitamin D deficiency is about 70-100% in general population worldwide. In India the increased prevalence of about 50-100% is due to low intake of dietary calcium [2, 3].

Vitamin D, a steroid hormone synthesized in the skin or acquired through diet, and parathyroid hormone (PTH), secreted by parathyroid glands, work synergistically to promote absorption of dietary calcium and phosphates [5]. Serum 25 hydroxyvitaminD [25(OH) D] is considered to be the best indicator of overall vitaminD status of an individual [4]. The active form of vitamin 3, 1, 25 dihydroxycholecalciferol [1,25(OH)₂D₃], functions within the calcium metabolic system to maintain serum calcium levels under homeostatic control. A sustained low 25(OH) D concentration, which limits production of 1, 25(OH)₂ D₃, ultimately leads to a slight decrease in serum calcium levels, triggering the secretion of PTH which stimulates bone resorption.

The medical fraternities across the world are curious in realizing that vitamin D plays a major role in health and disease. It causes skeletal as well as extra skeletal manifestations. Vitamin D not only regulates calcium and phosphorus homeostasis but also protects the individuals from many diseases like malignancies, chronic infections, and cardiac problems and reduces the risk of autoimmune diseases. The study is to impress upon the physicians about the gravity of the vitamin D deficiency problem throughout India and to make appropriate diagnosis and treatment with care and caution [6]. As the vitamin D status is improved worldwide it would have remarkable effects on public health, and decrease the healthcare expenses for several chronic diseases.

MATERIALS AND METHODS

This study was conducted in the Department of Biochemistry, Kilpauk Medical College Hospital, and Chennai. All procedures concerning human subjects or patients were permitted by the Institutional Ethical Committee. Explicit written consent was obtained from

the study population. The study population comprised of normal individuals coming for master health checkup Kilpauk Medical College Hospital, Chennai. Known smokers, alcoholics, diabetes, hypertensives, coronary artery disease were excluded from the study . 2ml of blood collected in plain serum tube for analysis of 25-OH vitamin D and PTH assay. 2ml of blood was collected in plain vials and serum was separated after centrifugation at 3000 rpm for 10 minutes and aliquoted, into an eppendorfs and stored at -20°C and were not thawed until the batch was analyzed for total calcium, phosphorus, total protein and albumin. All the biochemical analyses were performed using automated (ROBONIK) clinical chemistry analyser. All biochemical analyses were done using RANDOX calibrator (lot no 716UE) and Controls (lot no 919UN level 2, 723 UE level 3) for checking internal quality. The CVs of all the analytes performed were within the prescribed limits in accordance with CLIA.

Before storage ionized calcium is estimated. 25-OH vitamin D in serum is determined using ADVIA CENTAUR vitamin D assay on ADVIA CENTAUR XP systems. The ADVIA Centaur VitD assay is an 18-

minute antibody competitive immunoassay that uses an anti-fluorescein monoclonal mouse antibody covalently bound to paramagnetic particles (PMP), an anti-25(OH)vitamin D monoclonal mouse antibody labeled with acridinium ester (AE), and a vitamin D analog labeled with fluorescein. An inverse relationship exists between the amount of vitamin D present in the patient sample and the amount of relative light units (RLUs) detected by the system. The reference range of VIT D: 10–30 ng/ml.

The ADVIA Centaur Intact PTH assay is a two-site sandwich immunoassay uses direct chemiluminometric technology. It uses constant amounts of two anti-human PTH antibodies in the Lite Reagent. The polyclonal goat anti-human PTH (N-terminal 1-34) antibody labeled with acridinium ester is the first antibody. The biotinylated polyclonal goat anti-human PTH (39-84 regions) antibody is the second antibody. Streptavidin in the solid phase is covalently coupled to paramagnetic latex particles. The Reference range is 11.1 -79.5pg/ml

RESULTS

Table-1: Shows age, glucose, urea and creatinine among study population

Variable	Number	Mean	Std.dev
Age	100	38.2800	10.07154
Glucose	100	62.6000	22.96506
Urea	100	19.8200	6.08090
Creatinine	100	.8300	0.17321

This table shows the routine parameters (glucose, urea, and creatinine) within the study

population. And these parameters are within normal limits.

Table-2: Distribution of vitamin D levels in the study population.

Vit D	Group-1 (<10ng/ml)	Group-2 (10-20ng/ml)	Group-3 (>20ng/ml)
Numbers(n)	21 (21%)	43 (43%)	36 (36%)
Mean	6.947	14.31	30.392
Std dev	1.914	2.657	8.085

Based on the vitamin D levels the study population is grouped into three groups.

- Group-1 --- <10ng/ml (vit D deficiency)
- Group-2 ---- 10-20ng/ml (vit D insufficiency)
- Group-3 --- >20ng/ml (vit D sufficiency)

Among the study population about 21% of individuals are vitamin D deficient (group-1) and 43% individuals are vitamin D insufficient (group-2) and

36% of the population are vitamin D sufficient (group-3)

Table-3: Correlation of vitamin D with parathyroid hormone among the study population

	Vit D (ng/ml)	N	Mean	Std dev	p value
PTH	<10	21	68.3143	39.5684	0.002***
	10-20	43	50.0070	25.3879	
	>20	36	41.9211	16.9526	

Table- 3 shows correlation between vitamin D levels and parathyroid hormone.

A significant inverse relation was observed between vitamin D and intact parathyroid hormone levels in all the three groups. As vitamin D level

decreases there is a proportionate increase in parathormone level and is found to be statistically

significant with p value of 0.002.

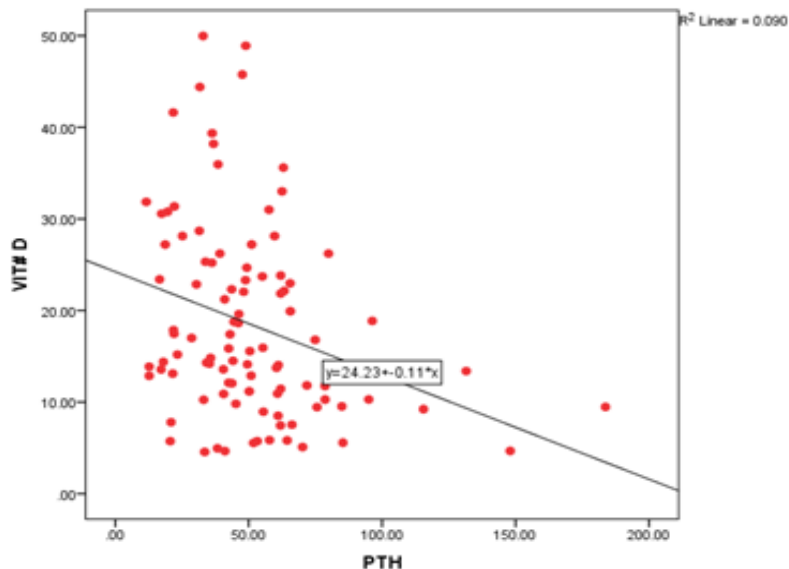


Table-4: Correlation of vitamin D with total calcium, ionized calcium, phosphorus

	Vit D	N	MEAN	STD DEV	p value
Total calcium	<10	21	8.146	1.03277	< 0.001
	10-20	43	9.2047	1.72157	
	>20	36	11.1778	1.09938	
I.calcium	<10	21	1.1048	0.08664	0.887
	10-20	43	1.1186	0.15001	
	>20	36	1.1056	0.13927	
Phosphorus	<10	21	3.6810	0.80164	0.337
	10-20	43	3.4605	0.57782	
	>20	36	3.4583	0.50787	

The total calcium in group 1 ----7.2- 9.2 mg/dl

In group 2 --- 7.5 – 10.9 mg/dl

In group 3 --- 10.1 -12.1mg/dl

In group 1 the total calcium level is decreased which is statistically significant with a p value <0.001. In group-2 and group-3 the total calcium is within normal limits and it is statistically insignificant.

The ionized calcium and phosphorus are found to be in normal range in all the three groups and it is also statistically insignificant with p value 0.887 and 0.337 respectively.

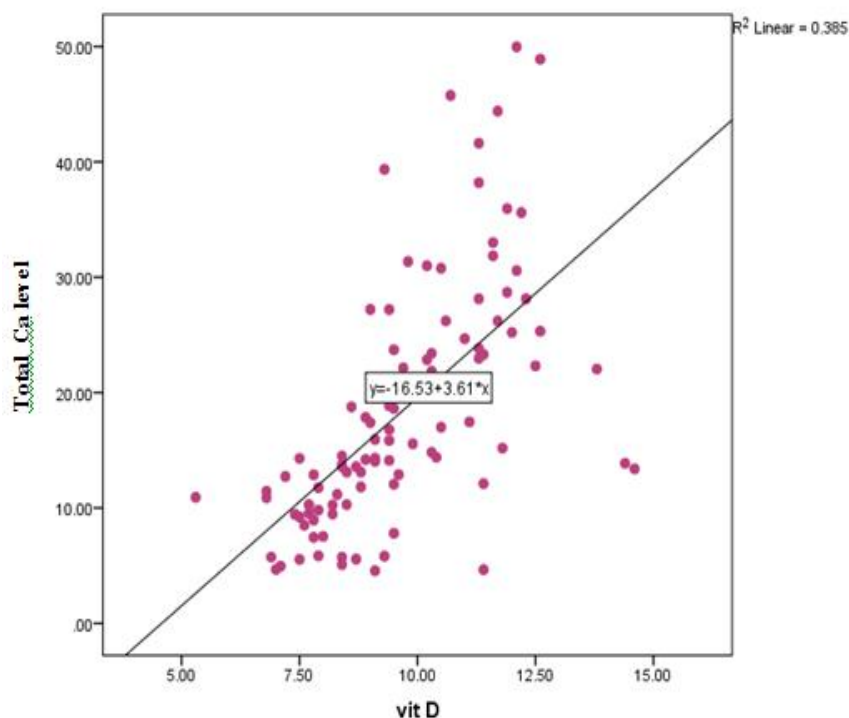
DISCUSSION

Assay of 25-OH vitamin D gives the best available measure of body stores of vitamin D and is the assay of choice for the diagnosis of vitamin D deficiency and vitamin D intoxication [13]. The present study is an attempt to evaluate the status of 25-OH vitaminD in unrelated normal healthy individuals in

Chennai population. Among the study population about 21% are found to be vitamin D deficient (<10ng/ml), 43% are vitamin D insufficient (10-20ng/ml) and 36% are vitamin D sufficient (>20ng/ml)

Correlation between vitamin D levels and parathyroid hormone, total calcium and ionized calcium were done in our study population and the results are as follows:

In this study the vitamin D levels have negative correlation with intact parathormone levels. As vitamin D level decreases there is a proportionate increase in parathormone level. Chapuy *et al.* in his study found a significant negative correlation between viamin D and intact PTH (p<0.01)[12].



The total calcium level was decreased in vitamin D deficiency individuals (<10ng/ml), whereas it was within normal range in other two groups.

Zuberi LM Habib A *et al.* stated in their study, calcium level is decreased only in severe vitamin D deficiency but it is normal in moderate and mild vitamin D deficiency which is similar to our study [7]. Mansoor. S. Habib *et al.* described that about 30% of asymptomatic adults with vitamin D deficiency and secondary hyperparathyroidism have normal levels of calcium [8].

Ionized calcium was within normal range in all the individuals of study population. In vitamin D deficiency, plasma calcium is maintained at the expense of bone calcium but persistence of deficiency leads to fall in calcium levels and secondary hyperparathyroidism [9]. Singh *et al.* reported that there exist a poor correlation between vitamin d deficiency and ionized calcium, so that vitamin D deficiency cannot be predicted by ionized calcium [10].

Serum phosphorus levels are within normal limits and it does not correlate with vitamin D levels in this study similar to the study conducted in Karachi. Revealed 30 to 40 % of calcium absorption in intestine is increased by vitamin D and phosphorus absorption by 80%. Vitamin D deficiency leads to secondary hyperparathyroidism which results in loss of phosphorus in urine and decrease the intestinal absorption of phosphorus. This results in low or low normal phosphorus concentration [11].

CONCLUSION

From this study we conclude that about 21% of individuals are vitamin D deficient. A significant inverse relation observed between 25(OH) D and intact parathyroid hormone. The total calcium level was decreased in vitamin D deficiency individuals (<10ng/ml). Ionized calcium and phosphorus was within normal range in all the individuals of study population and it does not correlate with vitamin D levels

Ethical approval

All procedures performed in these studies involving human participants were in accordance with the ethical standards of Institution.

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