

# Chemical Analysis of Vitamin D Insufficient and Vitamin D Deficient Patient in Dera Ghazi Khan

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## Abstract

**Background:** Among all vitamins, Vitamin D is important with respect to bone health and body immunity. Hypovitaminosis D is a global health issue, and considered as an epidemic. The worldwide, a billion of people with hypovitaminosis D has been estimated, however, there are a limited number of studies has been conducted in third world countries to measure the Vitamin D levels in their general population. This investigative study aimed to determine the prevalence of hypovitaminosis D in general population of District D. G. Khan, Pakistan.

**Methodology:** After a personal visit in various hospitals, 100 patients with vitamin D deficiency and insufficiency were enrolled in the study. They full filling inclusion and exclusion criteria. After taking informed written consent, blood samples were drawn and examine the serum levels of 25-hydroxy vitamin D. The serum level of 25-hydroxy vitamin D below 32 ng/ml is classified as Vitamin D insufficiency, while below 20 ng/ml is Vitamin D deficiency.

**Results:** The mean age of 62 male (18-50) years and 38 females (18-50) years. Among these, serum 25(OH) Vitamin D means level of 62 male samples while 38 female samples were studied. The 27 males showed vitamin D insufficiency, 25 males were Vitamin D deficient. Among 38 females, 11 subjects showed vitamin D insufficiency, 22 were vitamin D deficient, while 10 males and 5 females were normal control. The male subject showed significantly low serum calcium and phosphorous level, but elevated alkaline phosphatase (ALP) level as compared to normal male control. Similar finding has been found in female subjects.

**Conclusion:** The vitamin D deficiency or insufficiency reduces the serum calcium and phosphorous level, while it causes elevation of serum ALP level.

**Keywords:** 25-hydroxy vitamin D; Alkaline phosphatase; Calcium; Phosphorous

## Introduction

### Vitamin D

Fat-soluble vitamin D has two forms, vitamin D<sub>2</sub> and Vitamin D<sub>3</sub>. There are two main sources of vitamin D<sub>3</sub>, it is produced due to exposure of skin to UV (B) radiation or it is obtained through diet. While vitamin D<sub>2</sub>, found in a few plants also. Although both forms have same metabolic pathway, but it is found that vitamin D<sub>2</sub> more rapidly metabolize as compared to vitamin D<sub>3</sub>. Although Vitamin D is essential for intestinal absorption of calcium, it has been found that level of the 25(OH) D must be higher than 32 ng/mL for the optimal absorption of calcium [1,2].

## Materials and Method

### Field work

Patients were identified through personal visit in different areas of district D.G. Khan, the clinically diagnosed vitamin D insufficient and deficient patients were included in study. The physical symptoms were, i.e., fatigue, general muscle pain, weakness, joint pain, chronic pain, high blood pressure, restless sleep, headaches, urinary bladder problem, constipation and diarrhea. Preliminary information about the patients includes their contact number and postal address, etc.

Those patients were included in the study, which were diagnosed with vitamin D deficiency or insufficiency with no other reported disorder. The subjects were not included in the study if having; Anemia, genetic disorder, diabetes mellitus, cardiopulmonary disease, viral infection, bacterial infection and Malignancy [3-5].

### Collection of blood sample

The five ml of peripheral blood sample was collected from each patient by using butterfly needle and sterile syringes. The blood samples

were collected in plain tubes and tubes were allowed to stand for 30 min to blood clot, followed by centrifugation at 3000 rpm for 10 min to separate serum. Each serum sample was immediately transferred into serum collection tubes labeled with patient identification codes.

1. The vitamin D quantification was performed through the Vitamin D test kit.
2. The serum calcium level was quantified through photometric system.
3. The serum phosphorus level was measured spectrophotometric ally. For this purpose serum phosphorus quantification Diagnostic system kit.
4. The quantification of serum alkaline phosphatase (ALP) level was carried out through *in vitro* diagnostic reagent photometric system kit.

## Results

After a personal visit in various hospitals, 100 patients with vitamin D deficiency and insufficiency were enrolled in the study. They full filling inclusion and exclusion criteria. After taking informed written consent, blood samples were drawn and examine the serum levels of 25-hydroxy vitamin D. The serum level of 25-hydroxy vitamin D below

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32 ng/ml is classified as Vitamin D insufficiency, while below 20 ng/ml is Vitamin D deficiency [6].

The mean age of 63 male (18-50) years and 37 females (18-50) years. The 27 males showed vitamin insufficiency, 25 males were Vitamin D deficient among 38 females. 11 subjects showed vitamin D insufficiency, 22 were vitamin D deficient, while 10 males and 5 females were normal control. The male subject showed significantly low serum calcium and phosphorous level, but elevated alkaline phosphatase (ALP) level as compared to normal male control. Similar finding has been found in female subjects [7-9].

The current section provided the research findings regarding the responses of the patients with respect to the research questions of the study. These responses were analyze by using the frequencies as well as by applying the tools of t-test and ANOVA application to find out the responses rate on the research questions and to reach the conclusion comprehensively. The table provided the data regarding the frequencies of the respondents with respect to the gender. The gender demographic variable was grouped into males and females. The table shows that there were total 100 respondents who participate in the current research study. The table further shows that there were total 62 male respondents while the female respondents were 38 [10].

In Table 1, the male participants are 62.0% which shows significant participation of males instead of females.

The data regarding the frequencies of the respondents with respect to the age is given in Table 2. The age demographic variable was grouped into 20-35, 36-45 and 46-60. There were total 100 respondents who participate in the current research study. The table further shows that there were total 38 respondents who have age in between 20-35. While the female respondents those who have age in between 36-45 were 31. In the same line, the respondent with the age in between the 46-60 were also 31.

Three age groups were studied in present study with different cumulative %age.

Table 3 showing the vitamin D frequencies amongst the normal, deficient and insufficient participants.

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	Male	62	62.0	62.0	62.0
	Female	38	38.0	38.0	100.0
	Total	100	100.0	100.0	

Table 1: The frequencies regarding the gender.

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	20-35	38	38.0	38.0	38.0
	36-45	31	31.0	31.0	69.0
	46-60	31	31.0	31.0	100.0
	Total	100	100.0	100.0	

Table 2: The frequencies regarding the age.

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	Normal	15	15.0	15.0	15.0
	Deficient	38	38.0	38.0	53.0
	Insufficient	47	47.0	47.0	100.0
	Total	100	100.0	100.0	

Table 3: The frequencies regarding the vitamin D.

Table 4 showing the calcium frequencies amongst the normal, deficient and insufficient participants.

Table 5 showing phosphorous frequencies amongst the normal, deficient and insufficient participants.

Table 6 showing alkaline phosphatase frequencies amongst the normal, deficient and insufficient participants.

The data regarding the research questions given in Table 7 shows that the patients have different mean values of age, vitamin D, calcium, phosphorus and alkaline phosphatase. Table 7 provided the data with respect to the total number (100), the mean and the standard deviation. The research question shows the insignificance with respect to the gender based group differences.

The data regarding the demographic group mean differences with respect to gender given in Table 8. The table above shows that the

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	Normal	15	15.0	15.0	15.0
	Deficient	38	38.0	38.0	53.0
	Insufficient	47	47.0	47.0	100.0
	Total	100	100.0	100.0	

Table 4: The frequencies regarding the calcium.

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	Normal	15	15.0	15.0	15.0
	Deficient	38	38.0	38.0	53.0
	Insufficient	47	47.0	47.0	100.0
	Total	100	100.0	100.0	

Table 5: The frequencies regarding the phosphorus.

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	Normal	15	15.0	15.0	15.0
	Deficient	38	38.0	38.0	53.0
	Insufficient	47	47.0	47.0	100.0
	Total	100	100.0	100.0	

Table 6: The frequencies regarding the alkaline phosphatase.

	N	Minimum	Maximum	Mean	Std. Deviation
Gender	100	1	2	1.37	0.485
Age	100	1	3	1.93	0.832
Vitamin D	100	1	3	2.32	0.723
Calcium	100	1	3	2.32	0.723
Phosphorus	100	1	3	2.32	0.723
Alkaline Phosphatase	100	1	3	2.32	0.723
Valid N (list wise)	100				

Table 7: The descriptive statistics regarding the research questions.

	Gender	N	Mean	Std. Deviation	F	Sig.
Vitamin D	Male	63	2.24	0.712		
	Female	37	2.46	0.730	0.270	0.605
Calcium	Male	63	2.24	0.712		
	Female	37	2.46	0.730	0.270	0.605
Phosphorus	Male	63	2.24	0.712		
	Female	37	2.46	0.730	0.270	0.605
Alkaline Phosphatase	Male	63	2.24	0.712		
	Female	37	2.46	0.730	0.270	0.605

Table 8: The group statistics regarding gender-based mean differences (T-test).

patients have similar opinions regarding all the research questions and none of the research variable shown its significance with respect to gender. The table above provided the data with respect to the total number (100), the mean and the standard deviation. The research question shows the insignificance with respect to the gender based group differences [11,12].

## Discussion

Vitamin D is produced by the skin exposed to direct sunshine and is also provided by the nutrients. Each of these two sources can provide a sufficient supply, but if one of them is reduced the other source can easily become insufficient. Nevertheless sun effect even if the role of skin synthesis as a determinant of serum 25(OH) D has not been completely defined because of the absence of a direct measurement of personal sunshine exposure [13].

Our findings show that women were more likely to be vitamin D deficient than men. Vitamin D deficiency is often seen in post-menopausal women and has been associated with a greater incidence of hip fractures [14].

In our study, the influence of gender is reported in insufficiency, deficiency of vitamin D in serum, female is more affected (67%) as compared to male (37%). Our results are in accordance with [15-17]. Cultural dress (e.g. burkhas) is potentially a major contributor to reduced vitamin D<sub>3</sub> concentrations as the majority of the Asian population studied are of Pakistani origin. Since female Asians cover the majority of their bodies (e.g. purdah) this in turn will reduce the exposure to sunlight all year round and thus vitamin D<sub>3</sub> production [18]. Vitamin D<sub>3</sub> levels will therefore be depleted as an estimated 90% of the vitamin is produced *in vivo* unless replaced through diet or supplementation [19].

Vitamin D is a fundamentally important hormone involved in calcium absorption, bone mineralization and parathyroid hormone production. Vitamin D insufficiency and deficiency is involved in calcium and phosphorus absorption in the serum and availability in food, diet and intake. There is low calcium and phosphorus level associated with insufficiency and deficiency as proposed by Willis et al. [18].

In this study, the result of ANOVA shows highly significance with the role of alkaline phosphate level of serum and vitamin D in sufficiency and deficiency.

## Conclusion

In conclusion, we found that the vitamin D deficiency and insufficiency is associated with the levels of Calcium, Phosphate, Alkaline phosphate and gender and age of suspects. If normal levels of Calcium, phosphate and normal functioning of alkaline phosphate enzyme may lead to normal vitamin D level which is the overall health of bones and skeleton and body.

## Facts and Findings

I made recommendation for future studies based on our study findings and needs of D. G. Khan population. As well, I suggested some possible programs to increase Vitamin D level among D.G. Khan population living in south Punjab and perhaps to some extents provides to awareness to the Punjab health authorities of the Vitamin D status of their populations and if possible encourage them to take positive step forward to improve their populations health.

## Recommendation

The following preventive strategies are made for avoiding vitamin

D Insufficiency and Deficiency. The vitamin D Insufficient and Deficient patients should be used proper amount of foods and those foods which contain proper amount of vitamin D such as Code liver oil, Swordfish, Salmon, Orange juice, Milk, Margarine, Sardine, Liver, Beef, Eggs, Cereals. The major source of Vitamin D for children and adults is exposure to natural sunlight.

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