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Association of Vitamin D status with testosterone in people from north Gujarat, India

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ABSTRACT
The main aim of the present work is to study the association between vitamin D status with testosterone in healthy people from north Gujarat.This study represents a 2.25 years retrospective study involving reviewing the records of healthy Indians from north Gujarat. The 1109 subjects over 20 years of age who had undergone a comprehensive medical health check-up including testosterone, lipid profile, serum 25(OH)D HbA1C, Iron, and Haemoglobin levels were included. Chi square test was used to find the association between different vitamin D and testosterone. Total 84.85% of all subjects were found with vitamin D deficiency, with only 15.15% of subjects being vitamin D sufficient. Our results have shown lack of association between 25(OH)Dstatus and serum testosterone. Further studies to confirm the effects of lifestyle, food habits, disease condition, physical activity and vitamin D supplementation is advised.
Keywords: *vitamin D, testosterone, healthy Indians, deficiency, serum 25(OH)D.*

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INTRODUCTION

Vitamin D is a fat soluble vitamin that has been known to help the body absorb and preserve calcium and phosphorous; both are vital for building bone health. Decrease in vitamin D total levels may be due toinsufficient exposure of sunlight, dietary deficiency, nephrotic syndrome. Most of the vitamin D in our body is synthesized cutaneously upon exposure to ultraviolet B radiation in the sunlight and the rest is absorbed from our diet. Vitamin D is hydroxylated in the liver into 25-hydroxyvitamin D (25(OH)D), which is the main circulating form in the blood. In the kidney, 25(OH)D is hydroxylatedfurther into the metabolically active form, 1,25-dihydroxyvitamin D ($1,25(OH)_2D$)[1,2].The cut-off values for an individual are based on 25(OH)D level in the blood[3].

Recent epidemiological studies have discovered relationships between vitamin D and autoimmune diseases, cardiovascular diseases, cancer, depression and diabetes mellitus[4,5].Testosterone is a crucial steroidal hormone produced by both men and women. In men, testosterone levels raise during puberty and stimulate the production of sperm and the development of secondary sexual characteristics like body hair and a deeper voice. Testosterone help tomaintain bone strength, stimulates the development of muscle mass and strength, and improves libido and mood in both men and women [6].In the beginning, vitamin D hormones were believed to be limited to the liver and kidney but have recently been observed in other tissues also. Particularly, vitamin D metabolising enzymes and receptors have been identified in the testes, demonstrating that vitamin D may have a role in regulating testosterone production[7].

Cross-sectional studies establishing the relationship between vitamin D and testosterone have revealed heterogeneous results. Even though some studies have shown a positive relationship between them,[8,9]others have showed on-significant relationship[10]. Similarly, studies on the effects of vitamin D supplementation on testosterone levels have yielded diverged results. The study by Pilz et al. has shown that vitamin D supplementation increases testosterone level, but the outcomes by Jorde et al. are negative[11,12].There is little current data on the association between vitamin D levels and testosterone available from Asia. A Korean study demonstrated higher level of 25(OH)D was associated with higher total testosterone and free testosterone levels in Korean men[13].Here, we present results on association between serum testosterone concentrations and vitamin D3 deficiency among healthy participants.

MATERIAL AND METHODS

This study represents a 2.25 years retrospective study (January 2021 to March 2022) involving reviewing the records of healthy Indians from north Gujarat.

We included the subjects over 20 years of age who had undergone a comprehensive medical health check-up including testosterone, lipid profile, serum 25(OH)D HbA1C, Iron, and Haemoglobin levels. All records were evaluated for demographics detail, the exclusion criteria were presence of any health conditions. The study protocol was approved by the Sarvajanik Clinical Research Ethics Committee (DCGI reg. No. :ECR/295/Indt/GJ/2018).

The laboratory reports indicated that basal blood samples for 25(OH)D and testosterone were collected after an overnight fast. Levels of 25(OH)D measured by immunoassays were used for evaluation of inclusion criteria. According to recent clinical guidelines, vitamin D deficiency was defined as a serum 25(OH)D level < 30 ng ml^{-1}[14].To determine the testosterone status, venous blood samples were collected after a overnight fasting and measured using a competitive chemiluminescence immunoassay procedure. According to clinical guidelines Testosterone deficiency was defined as a serum level shown in Table-1[15].

Table1. Normal values of testosterone level

Age	Limits of Testosterone level (ng/dL)	
	Adult male	Adult female
21-49 year	164.94-753.38	-
50-89 year	86.49-788.22	-
Pre-menopause	-	12.09-59.46
Post-menopause	-	7.00-48.93

Data was collected using Microsoft excel which contain different continuous and categorical variables. Continuous variables are represented using mean and standard deviation. Categorical variables are represented using frequency and percentage also pie diagram and bar graph are used to present the frequency. Chi square test is used to find the association between different categorical variables.All statistical procedures were performed with SPSS version 20[16].$\boldsymbol{P} < 0.05$ was considered statistically significant.

RESULTS AND DISCUSSION

A total of 584 male and 525 female subjects aged >20 years were included in this study. Their mean age was 46.97± 13.39 years. The mean serum 25(OH)D concentration was found to be22.48± 10.80 ng ml^{-1}(Table-2).

Table 2. Age and Vitamin D status range

Variables	Minimum	Maximum	Mean	Std. Deviation
Age (years)	21	94	46.97	13.39
25(OH)D (ng/ml)	6	97	22.48	10.803

Surprisingly, 84.85% of all subjects were vitamin D deficient [25(OH)D <30 ng ml^{-1}], with only 15.15% of subjects being vitamin D sufficient [25(OH)D ≥ 30 ng ml^{-1}]. The overall proportion of deficiency level remained same in all age groups of adults (Table 3).

Table 3. Age-group wise distribution of vitamin D status

25(OH)D status	Age groups (years)			Total
	21 to 40	41 to 60	>60	
Deficient (<30 ng/ml)	333	431	177	941
Sufficient (30-100 ng/ml)	49	91	28	168
Total	382	522	205	1109

Several studies of vitamin D status have concluded high vitamin D deficiency around the world.Serum 25(OH)D concentrations of less than 20 ngl/ml are present in nearby37% of the worldwide population[17].Severe vitamin D deficiency prevails inabout18% of the US population,40% of Europeans, and a greater percentage of people living in the Middle East or Gulf states[18, 19].Severe vitamin D

deficiency has also been reported from Northern India and China[20].Our results indicates the extensive prevalence of Vitamin D deficiency, reported in the world so far. The prevalence of vitamin D deficiency was found to be almost same between the men and women participants (Table 4).

Table4. Gender wise prevalence of vitamin D deficiency

25(OH)D status	**Gender**		**Total**
	F	**M**	
Deficient (<30 ng/ml)	440	501	941
Sufficient (30-100 ng/ml)	85	83	168
Total	525	584	1109

Table 5. Associations of serum testosterone with vitamin D

25(OH)D status	**Testosterone status**		**Total**	**P#**
	Abnormal	Normal		
Deficient (<30 ng/ml)	424	517	941	0.126
Sufficient (30-100 ng/ml)	65	103	168	
Total	489	620	1109	

calculated by chi-square test

We performed review of data of serum testosterone level in total 1109 healthy subjects. It was found than more than 44 % subject were deficient for serum testosterone. Lower testosterone in vitamin D deficient subjects was observed but it was not significant ($p>0.05$). Similar to our results previous studies carried out on university students and health men have shown a non-association between 25(OH)D status and testosterone level[21-24].Results contrary to ours were obtained by various groups, showing a statistically significant association between 25(OH)D level and total and free testosterone level[25-29].

CONCLUSIONS

In conclusion, the results of our study demonstrate the absence of an association between deficient 25(OH)D concentration and testosterone levels in a group of healthy subjects from the north Gujarat, India. Limitation of our study is that it involves the people those are health conscious and visit the laboratory/hospitals for routine heath check-up, it does not represent the whole population. Therefore, more studies to verify the effects of lifestyle, food habits, disease condition, physical activity and vitamin D supplementation would be helpful.

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