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Research Article

A PILOT STUDY ON VITAMIN-D STATUS AND METABOLIC SYNDROME IN ADULT INDIAN POPULATION

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Abstract

Vitamin D status indicated by 25-hydroxyvitamin D, along with its classic role in calcium homeostasis and bone metabolism, is thought to be inversely associated with adiposity, glucose homeostasis, lipid profiles, and blood pressure. Studies among the western population have also shown it to be inversely associated with metabolic syndrome (MS) and cardiovascular diseases (CVDs). However, evidence from the Asian population is limited. Therefore, the present study aimed to study the prevalence of vitamin D deficiency (VDD) (<20 ng/mL) and its association with metabolic risk factors among adults. About 129 adults in the age group 30–60 years, who gave consent for participation, were enrolled in the study through snowball effect. Anthropometric measurements were recorded using standard methods & tools. Serum 25(OH)D was estimated by CLIA method, lipid profile and fasting-glucose using enzymatic kits. MS was defined using NCEP ATP-III, 2005 guidelines. The results revealed that, around 88% of the subjects had VDD, with the prevalence being significantly higher among females than males (95% vs 77%). 31% of the population had MS of which 87.5% were vitamin-D deficient. The vitamin-D status of the subjects was not significantly related with the risk of developing MS (OR=0.88, 95% CI= 0.28-2.78). None of the risk factors for MS correlated significantly with the vitamin-D levels of the subjects. Waist-circumference (Exp(B)= 0.281, 95% CI= 0.089-0.887) entered the model for logistic regression with vitamin-D status as the dependent variable. Thus the results highlighted the high prevalence of VDD among the population and it was independently associated with greater abdominal obesity.

Key words: Vitamin-D deficiency; metabolic syndrome; prevalence; adults; India

Introduction

Vitamin D deficiency has assumed pandemic proportions globally. Using serum (25[OH]D) values; a generally accepted indicator of vitamin-D status, it has been estimated that 1 billion people worldwide are vitamin-D deficient or insufficient (Holick, 2007). Studies conducted in India among various socio-economic groups, different ages, and both genders as well as different disease states, such as primary hyperparathyroidism report widespread vitamin D deficiency / insufficiency in the country (Harinarayan & Joshi, 2009; Harinarayan *et al*, 1995). The prevalence ranges from 70-90% (Goswami *et al*, 2000; Harinarayan *et al*, 2011; Marwaha *et al*, 2011). The individual's behaviour to avoid sunlight or clothing which prevents exposure to sunlight and low availability as well as intake of vitamin-D rich foods are thought to be the probable reasons.

Evidence suggests that apart from its classic role in calcium homeostasis and bone metabolism, vitamin-D status is associated inversely with blood pressure, adiposity, lipid parameters and glucose metabolism (Holick, 2007;

Wortsman *et al*, 2000; Chiu *et al*, 2004; Scragg *et al*, 2004; Hypponen *et al*, 2008; Pittas *et al*, 2007) through direct modulation of gene expression via vitamin-D receptors (VDRs) (Holick, 2007). Parkera *et al* (2010) reported that individuals with highest levels of serum vitamin-D were associated with a 43% reduction in cardio-metabolic disorders (OR 0.57, 95% CI: 0.48-0.68).

Epidemiological studies from western population suggest that vitamin-D status is inversely associated with metabolic syndrome which is a constellation of cardiometabolic risk factors (Forouhi *et al*, 2008; Hypponen *et al* 2008; Ford *et al*, 2005; Reis *et al*, 2008). Regarding this evidence from the Asian population is limited and as vitamin-D metabolism is affected by ethnicity and dietary habits, it is rather difficult to extrapolate the findings of western population to Asians (Chiu *et al*, 2004). Rapid nutrition and lifestyle transitions in the past few years have resulted in metabolic syndrome emerging as one of the most widespread health problems in Asian countries (Cornier *et al*, 2008). However, much remains to be learned about the relationship between vitamin-D status and metabolic

syndrome among Asian individuals. Therefore, the objective of our study was to demonstrate the association between serum 25(OH)D & metabolic risk factors and to identify the determinants of metabolic syndrome against the vitamin-D status in adult population of western India.

Methods

Selection and Description of Participants

This is a cross-sectional study conducted in an urban setting. Vadodara city in the state of Gujarat, India was divided into five zones. One society was selected purposively for the enrolment of the subjects in the age group 30-60 years. Then through snow ball effect the individuals were included till the required sample size was attained. Based on the available literature taking the prevalence of vitamin D deficiency as 80% in the adult Indian population, the sample size came out to be 138. A total of 141 participants (males=50 and females =91) were enrolled between the months of February to August 2013. For this particular research the participants with confirmed diabetes mellitus (n=12) were excluded for the analysis, as they are more probable to have metabolic syndrome. Hence the final sample size used was 129 participants (males=47; females=82).

Ethics considerations

The study was approved by the *Institutional Ethics Committee for Human Research* (Reference No. IECHR/2012/21) which is responsible for ethical issues in all research projects involving humans conducted by the department. Written informed consent was taken from all the participants.

Data Collection

Information regarding socio-demographic characteristics, medical history of hypertension (self-reported as diagnosed by a doctor) was collected using a pre-tested semi-structured questionnaire. Anthropometric measurements were also recorded. Weight was taken using calibrated Salter electronic bathroom scale. Height, waist and hip circumference were measured using non-stretchable fibre glass tape to the nearest 0.1cm. The waist was defined as the point midway between the iliac crest and coastal margin; while hip circumference as the widest circumference over the buttocks and below the iliac crest (WHO, 1995; National Institutes of Health, 1998). All measurements were taken by trained researchers. Body mass index (BMI) was calculated by standard formula of weight in kg / height² in meters. Overweight was defined as BMI range of 23-24.9 kgm² and obese as BMI \geq 25 (WHO, 2004). Metabolic Syndrome was defined as per the modified National Cholesterol Education Program Adult Treatment Panel III criteria (Grundy *et al*, 2005), where the presence of any three of the below five factors is taken for diagnosis of MS: abdominal obesity (waist circumference > 90 cm in men and > 80 cm in women), hypertriglyceridaemia (triglycerides \geq 150 mg/dL); low HDL cholesterol (HDL cholesterol \leq 40 mg/dL for men and \leq 50 mg/dL for women); elevated blood

pressure (systolic blood pressure \geq 130 mmHg and/or diastolic blood pressure \geq 85 mmHg or current use of antihypertensive drugs); impaired fasting glucose (fasting plasma glucose \geq 100 mg/dL).

Blood pressure was measured by clinically validated digital BP meter (Omron HEM-7203 model). For the biochemical profile, after an over-night fast, venous blood samples were collected by a trained technician. 25-hydroxyvitamin D for vitamin D status was estimated by Chemiluminescence Immunoassay (CLIA) technique in an accredited laboratory. The serum concentration of 25(OH)D is widely regarded as a "gold standard" indicator of vitamin D status as it is a good reflection of cumulative exposure to sunlight and dietary intake of vitamin D (Springbett *et al*, 2010). VDD was defined as serum 25(OH)D concentration of <20ng/ml and categorized as insufficiency (20- \leq 30 ng/ml) and sufficiency (>30 ng/ml) (Lips, 2001). Vitamin-D is reported in two units ng/ml & nmol/L (to convert ng/ml to nmol/L multiple the previous by 2.5). Lipid profile and fasting blood sugar was estimated using enzymatic kits. These biochemical analyses were outsourced to a private laboratory certified by ISO 9001:2008, NABL (India) and CAP (College of American Pathologist).

Statistics

Data was entered in Microsoft Excel 2007 and verified by the researcher. Statistical Package for Social Sciences (SPSS) version 16 and EpiInfo version 7 were used for statistical analysis of the data. A two-tailed p value was used for calculating statistical significance; a value of p<0.05 was taken to be significant. For quantifiable variables, descriptive statistics (means and standard deviation) were calculated. In case of categorical variables, frequency distribution was computed. Comparison of means was done using independent t-test and difference in proportions was compared by chi-square test. Pearson's correlation was performed to study the relationship between the various risk factors for MS and vitamin-D levels. Multiple logistic regression was conducted to predict the risk factors significantly associated with vitamin-D deficiency.

Results

A total of 129 individuals participated in the study of which 63.5% were females. The mean age of participants was 44.5 \pm 9.1 years (Table 1). Fifteen participants reported presence of hypertension and were on anti-hypertensive drugs. Around 70% of the participants were overweight or obese with males having significant higher prevalence for obesity. 31% participants had metabolic syndrome with no significant difference among the genders. Only five individuals had serum 25(OH)D levels in the sufficiency range (>30 ng/ml) while majority of them (88.4 %) had vitamin-D deficiency, with significantly higher rates among females (p<0.01). Table 1 also shows the gender wise metabolic risk factors among the participants. No significant difference was observed for the systolic blood

pressure among the participants while males had significantly higher levels of diastolic blood pressure (83.6 ± 10.6 vs 79.2 ± 9.8 , $p < 0.05$). Female participants showed significantly higher readings for both the bad LDL-cholesterol ($p < 0.01$) and the good HDL-cholesterol ($p < 0.001$), while lower levels for serum vitamin D ($p < 0.001$) than their male counterparts. There was no difference observed for rest of the risk factors among both the genders.

The Odds Ratio (OR) of age, gender, presence of MS and the metabolic risk factors with vitamin-D status is depicted in table 2. Female gender (OR= 5.95; 95% CI 1.77-19.99,

$p < 0.01$) and waist circumference (OR= 3.55; 95% CI 1.1-11.2, $p < 0.05$) had significantly higher odds for having vitamin-D deficiency (< 20 ng/ml). Age and metabolic risk factors were not statistically associated with vitamin-D status. Also the vitamin-D status of the subjects was not significantly related with the risk of developing MS (OR=0.88, 95% CI= 0.28-2.78). A correlation was studied between the vitamin-D status of the participants and the risk factors for MS (Table 3). It was observed that except for blood pressure, rest of the risk factors showed a negative non-significant correlation with the vitamin-D status of the participants.

Table 1: Gender wise baseline characteristics of the participants

Characteristic	Males (n=47)	Females (n=82) n, % #	Total (n=129)	p-value
Hypertension (self-reported)	3 (6.4)	12 (14.6)	15 (11.6)	0.253
BMI Status				0.041*
Normal	13 (27.6)	26 (31.7)	39 (30.2)	
Overweight	5 (10.6)	22 (26.8)	27 (20.9)	
Obese	29 (61.7)	34 (41.5)	63 (48.8)	
25(OH)D deficiency (<20 ng/ml)	36 (76.6)	78 (95.1)	114 (88.4)	0.005**
Metabolic syndrome	16 (34)	24 (29.3)	40 (31)	0.573
		Mean \pm S \$		
Age (years)	43.5 \pm 8.4	45.1 \pm 9.5	44.5 \pm 9.1	0.336
BMI (kg/m ²)	26.5 \pm 4.7	25.5 \pm 4.7	25.8 \pm 4.7	0.288
Waist circumference (cm)	92.5 \pm 11.1	92.8 \pm 10.9	92.5 \pm 11.1	0.580
Systolic BP (mm Hg)	132.2 \pm 16.3	126.7 \pm 16.8	128.7 \pm 16.7	0.073
Diastolic BP (mm Hg)	83.6 \pm 10.6	79.2 \pm 9.8	80.8 \pm 10.4	0.020*
Fasting Blood Sugar (mg%)	85.0 \pm 12.4	84.2 \pm 17.1	84.5 \pm 15.5	0.779
Total cholesterol (mg%)	186.8 \pm 29.6	194.7 \pm 36.6	191.9 \pm 34.3	0.211
LDL-Cholesterol (mg%)	102.3 \pm 21.6	116.3 \pm 31.2	111.1 \pm 28.8	0.003**
HDL-Cholesterol (mg%)	45.1 \pm 8.7	56.1 \pm 12.5	52.1 \pm 12.4	0.000***
Triglycerides (mg%)	137.7 \pm 89.7	112.4 \pm 49.4	121.6 \pm 67.7	0.079
25(OH)D (ng/ml)	17.6 \pm 5.5	11.4 \pm 7.1	13.7 \pm 7.2	0.000***

chi-square test, \$ independent t-test. Values in parenthesis are in percent. $p < 0.001$ ***, < 0.01 **, < 0.05 *

Table 2: Odds Ratio for factors of age, gender and metabolic factors vs vitamin-D status

Risk Factor	25(OH)D <20 ng/ml (n=114)	25(OH)D ≥ 20 ng/ml (n=15)	OR	95% CI
Age (≥ 40 years)	74 (64.9)	12 (80)	0.46	0.12-1.73
Gender (female)	78 (68.4)	4 (26.7)	5.95**	1.77-19.99
WC (M ≥ 90 , F ≥ 80)	84.2 (96)	60 (9)	3.55*	1.1-11.2
SBP (> 130 mmHg)	41.2 (47)	20 (3)	2.80	0.7-10.5
DBP (> 85 mmHg)	28.1 (32)	26.7 (4)	1.07	0.3-3.6
TG (≥ 150 mg%)	21.1 (24)	6.7 (1)	3.73	0.5-29.8
HDL-C (M < 40 , F < 50)	42.1 (48)	40 (6)	0.91	0.3-2.7
FBS (> 100 mg%)	11.4 (13)	6.7 (1)	1.80	0.2-14.8
Metabolic syndrome	35 (30.7)	5 (33.3)	0.88	0.28-2.78

Values in parenthesis are in percent; $p < 0.01$ **, < 0.05 *

Table 3: Correlation between vitamin-D levels and risk factors of Metabolic Syndrome

Risk factor	Pearson's correlation (r)	p-value
Abdominal Obesity	-0.024	0.784
High Blood Pressure	0.032	0.721
Hyperglycemia	-0.127	0.151
Low HDL-Cholesterol	-0.113	0.204
Hypertriglycerdemia	-0.02	0.823

Table 4: Logistic Regression for the determinants of MS with vitamin-D as dependent variable

		OR [Exp(B)]	p-value	95% CI
Variable in equation	Waist circumference	0.281	0.03*	0.089-0.887
Variables not in equation	Systolic BP		0.154	
	Diastolic BP		0.883	
	HDL Cholesterol		0.663	
	Triglyceride		0.196	
	Fasting blood sugar		0.626	

p <0.05*

A multivariate model was developed to identify the determinants of MS with vitamin-D as the dependent variable (Table 4). Only waist-circumference (Exp(B)=0.281, 95% CI= 0.089-0.887) entered this model of logistic regression thus suggesting that low vitamin-D levels were independently associated with abdominal obesity.

Discussions

The present study demonstrated the vitamin-D status of the participants based on their serum 25(OH)D levels. A large proportion of the participants (approximately 88%) were vitamin-D deficient with levels below 20 ng/ml. In a recent review the prevalence of various countries throughout all continents, ranged from 31% in Australia to 98% in Mongolia. The reported prevalence for Indian population was 75% (Wacker & Holick, 2013). Enough information is not available on vitamin-D status of population in Asian region, except few studies conducted among children, pregnant women or post-menopausal women and elderly population. In Beijing, China the geometric mean plasma 25(OH)D was 40.4 nmol/L and prevalence of vitamin-D deficiency was 69.2% among middle-aged and elderly individuals (Lu *et al*, 2009), while in Japan low serum levels (34 nmol/L) were observed in women younger than 30 years and in immobile older persons (30 nmol/L) (Nakamura *et al*, 2001). In Korean population also vitamin-D insufficiency (<50 nmol/L) was found in 47.3% males and 64.5% females in the Korea national health and Nutrition Examination Survey (KNHANES) (Choi *et al*, 2008). Despite being a sunny country, India has not been spared of low serum vitamin-D levels. In a survey among hospital staff the mean serum levels were 30 nmol/L (Arya *et al*, 2004), while among pregnant women it was 35 nmol/L (Sachan *et al*, 2005) and 36 nmol/L for the post-menopausal women in India (Harinarayan, 2005). Similar low levels (13.7 ng/ml or 34.2 nmol/L) have been reported in our study also. Our female participants had significantly lower mean 25(OH)D levels than males, which is similar to studies reported elsewhere (Zargar *et al*, 2007; Rahnavard *et al*, 2010; Lips, 2010; Moy & Bulgiba, 2011). The clothing style (wearing long sleeves salwar-kameez or sari) and long inactive indoor hours due to household work could be the possible reasons contributing to this difference.

Based on BMI, a high prevalence of overweight and obesity was seen in our population with mean BMI of 26.5 ± 4.7 . This may also be a probable reason for low vitamin-D levels as reported by Worstman *et al* (2000) who in his study showed that high BMI leads to a lower vitamin-D concentration because vitamin-D is a fat soluble vitamin that is not easily released from adipose tissue once absorbed among obese individuals. Abdominal obesity was significantly associated with low vitamin-D levels in our study; similar to results reported in other studies (Moy & Bulgiba, 2011; Lu *et al*, 2009; Liu *et al*, 2005; McGill *et al*, 2008). Consequently this obese population would be more susceptible to metabolic syndrome (Ford *et al*, 2005) as well as cardiovascular disease (Grandi *et al*, 2010; Zittermann & Gummert, 2010; Wang *et al*, 2008).

The relationship between metabolic syndrome and vitamin-D is still unclear. In our study, about one-third of the population had metabolic syndrome (atleast 3 of the 5 risk factors), but their vitamin-D status was not significantly related with the risk of developing it. Among the various metabolic risk factors versus vitamin-D status, significant associations were observed only for female gender (p<0.01) and waist circumference (p<0.05). Individuals with low vitamin-D levels had higher but non-significant odds for high systolic BP, hypertriglyceridemia and hyperglycemia. As reported elsewhere (Parkera *et al*, 2010; Kim *et al*, 2010, Moy & Bulgiba, 2011; Ford *et al*, 2009), vitamin-D insufficiency or deficiency was independently associated with increased risk of having metabolic syndrome. Our study also investigated the correlation between metabolic risk factors and vitamin-D levels; however none of the risk factor showed a significant relationship. Except for blood pressure, a negative correlation was observed for the remaining risk factors. Our findings are contrary to the ones reported by other authors (Kim *et al*, 2013; McGill *et al*, 2008). The small sample size of our study may not have adequate power to detect significant association and causative relationship between vitamin-D status and individual risk factors. Multivariable regression analysis revealed that abdominal obesity (p<0.05) was independently associated with poor vitamin-D status in our study, which again emphasises the relationship between high fat accumulation in the body and low vitamin-D levels in the serum.

The limitations of our study include the cross-sectional design where the cause could not be attributed and the small sample size with inadequate power, which would have failed to identify the association of vitamin-D status with some of the risk factors for metabolic syndrome. This study was based on a single measurement of serum 25(OH)D as an indicator of vitamin-D status and parathyroid hormone which is a functional index of vitamin-D status could also not be measured due to monetary constraints. The results cannot be generalised to entire nation as our participants belonged only to the urban setting. However, to the best of our knowledge, this is the first pilot study to investigate the association of 25(OH)D levels with metabolic syndrome in males and females of India more so in western India which receives ample sunlight for almost nine months of the year. This study is also timely as literature reports a high prevalence of vitamin-D deficiency in India, which supports the need for a nationalized supplementation programme. Our study will add to the knowledge of researchers and existing literature and aid in policy making.

Conclusions

Our findings have given rise to concerns about the vitamin-D status of the Indian population in general and its adverse cardio-metabolic effects on health. Hence it is recommended that the assessment of vitamin D status among the population, more particularly in females should be a regular feature in routine health check-ups. There is a felt need to frame studies focussing on diet and physical activity which will help in preventing abdominal obesity and in improving vitamin-D levels and thus correcting the metabolic alterations in the body.

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Declaration

A part of the data has been presented at the national conference on 'Nutrigenomics- A promising tool for combating chronic diseases' on 3-4 February 2014 at Sardar Patel University, Vallabh Vidyanagar, Gujarat. India.

The authors declare no conflict of interest.

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