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VITAMIN D STATUS AMONG NON-PREGNANT ADULT FEMALES: A STUDY FROM NORTH EAST INDIA

Clinical Research

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ABSTRACT

Vitamin D deficiency reported in epidemic proportions worldwide including India. There is a paucity of data regarding vitamin D insufficiency among north-eastern Indian adult females. The present study was conducted to look into vitamin D status among adult non-pregnant females of north-eastern India, and to find its association with risk factors. Ninety two non-pregnant ladies aged 18-52 years were included in the study. Serum 25 OH Vitamin D total level was measured by Enzyme linked fluorescent assay (ELFA) method. 97.8%.of the studied population were found to have vitamin D deficiency/insufficiency. The Vitamin D deficiency/insufficiency is found to be more prevalent in younger age groups and more so in unmarried women. There is a significant negative correlation was found between socioeconomic status and measured vitamin D (p=0.00). Serum Immunoglobulin E (IgE) level showed negative correlation trend with vitamin D. Study showed a high prevalence of vitamin D deficiency/insufficiency in the studied population similar to other parts of India.

KEYWORDS

Vitamin D, adult female, non-pregnant, north-east India, ELFA

INTRODUCTION

Vitamin D is a lipid-soluble steroid prohormone. By the action of solar ultraviolet B radiation, Vitamin D is synthesized from 7-dehydrocholesterol on the human skin. It also derived from dietary sources. Usually, 50–90% of required vitamin D is synthesised by the skin on exposure to sunlight. The biologically active form of Vitamin D is obtained within the human body after two successive hydroxylations in liver and kidney/other tissues. The 25-(OH) vitamin D is the chief storage form of vitamin D in the human body and therefore it is the preferred analyte to determine the vitamin D status[1].

Recent research highlighted the various physiological role of vitamin D beyond the well-established musculoskeletal functions and requirement in calcium homeostasis[2]. Extra-skeletal benefits are also supported by the universal presence of vitamin D receptors throughout the human body controlled by approximately 3000 genes[3]. Early life vitamin D insufficiency found to be associated with increased incidences of wheezing and respiratory infection in the children with subsequent chronic implications in adult life[4]. Diminished levels of vitamin D have also been associated with autoimmune thyroid disorders like Hashimoto's thyroiditis and Graves' disease. Compromised vitamin D signalling pathway has been postulated to be related to thyroid carcinogenesis[5]. Numerous publications established the link between the low circulating level of vitamin D and increasing incidences of autoimmune disorders, cardiovascular diseases, diabetes, and cancer. It has also been suggested that vitamin D-deficiency can adversely affect the females of reproductive age by increasing the risk for pre-eclampsia, pregnancy-induced hypertension, prolonged labour, and low birth weight of infant leading to adverse reproductive outcomes[6].

Vitamin D deficiency is a well-known global pandemic but it also recognized as the most poorly managed nutritional deficiency worldwide. The postulated prevalence of vitamin D is projected to affect more than one billion people throughout the world[7]. India is a tropical country with abundant sun exposure but countrywide several studies have reported that 70%–100% healthy individuals are deficient of vitamin D. Even then currently vitamin D insufficiency is neither declared as National Health Priority (NHP) nor included in any national health program[8]. In a study it was found 14%–32% of young healthy females from the Indian paramilitary forces were osteopenic though consumed a balanced diet, had adequate exposure to sunlight, and undertook regular outdoor physical activities[9]. A study in North India showed a very high prevalence (91%) of VDD among adult rural females[10].

There is scarce of research data regarding vitamin D insufficiency among north eastern Indian adult females, and specifically in the urban migrated pan Indian community. The present study was conducted to estimate the prevalence of 25-OH vitamin D deficiency/insufficiency among adult non-pregnant apparently healthy females residing in an urban community of North-Eastern India, and to find its association with various risk factors.

MATERIAL AND METHOD

This was a cross-sectional, community-based observational study. The study population consists of 92 non-pregnant adult apparently healthy ladies who had attended a one-day health camp of 699 bedded hospital of Guwahati, Assam. All the study participants were from middle-class socioeconomic strata with more than basic educational level. Though all of them are ethnically Indian, study participants were from different parts of the country and now residing in the north-eastern part of the country due to occupational/ educational reasons. The participants are living in the city area with adequate space for outdoor physical activity and away of general pollution of the city. The study conducted in the spring season in the month of March which is the most pleasant weather in the region for outdoor activities and has a probability to maximum exposure to sunlight. All the study participants were covered under organised scientific healthy diet ration services for their daily household need. They have good access to free basic health care facilities. In the past 7 days, participants who consumed satisfactory Vitamin D rich food like milk or dairy products, fish, egg yolk etc. for more than 3 days within last 01 week period were considered to have a satisfactory diet .Questions were asked regarding Vitamin D supplementation within a month period. All the ladies who attended the clinic were evaluated for short clinical history for acute disease and chronic medications by a competent gynaecologist. The study population was divided into 10 socio economic strata based on the income range of head of family. In our study based on the interview, we presumed that the study population had adequate facilities for exposure of natural sources of vitamin D. Written informed consent was obtained from the participants. Ethical approval for the study was obtained from the Ethical Committee of the institution. Pulse and blood pressure were measured with electronic apparatus and subsequently fasting blood samples were collected in appropriate EDTA, sodium fluoride and plain vaccutainer respectively for evaluation of complete blood count, fasting blood sugar, serum Thyroid stimulating hormone (TSH), Serum Immunoglobulin E (IgE) and serum 25 OH Vitamin D total level. Complete blood count was done in 03 parts haematoanalyser. Fasting blood sugar was measured by advanced adapted hexokinase-glucose-6-phosphate dehydrogenase method in fully automated biochemistry analyser. Serum TSH, total IgE and 25 OH Vitamin D total were assessed by competitive enzyme immunoassay with a final fluorescent detection or

(ELFA) method in fully automated immunoanalyser with proper quality control. Samples collected were stored in the primary tube at room temperature and were processed within 04 hours of sample collection.

Blood haemoglobin concentration < 12 gm/dl was taken as defining criteria for anaemia for adult non-pregnant females according to the World Health Organization (WHO) definition. Following the 2018 ADA criteria, fasting blood sugar level < 100mg/dl was taken as normal and sugar level > 126mg/dl is defined as the diabetic range. Sugar level between 100 and 125 mg/dl was considered as pre-diabetic impaired fasting range abnormality[11]. The analytical reference range of the sugar detection method was 0-500mg/dl.

Serum TSH and serum total IgE reference range were taken as 0.25 – 5 µIU/ml and <150KIU/l as per kit literature. Method-specific analytical detection limit for serum total IgE was 0.5 KIU/l. The intra and interassay coefficient of variation for serum total IgE assay were 4.5% and 3.7% for low serum range and 3.7% and 5% for high serum range, respectively. The measurement range of the TSH kit used was spread up to 60 µIU/ml and the analytical detection limit was 0.05 µIU/ml. The intra and inter assay coefficient of variation for utilised serum TSH assay was 4.7% and 3.5% for low serum range and 2.4% and 3.2% for high serum range, respectively. In our study, serum 25 OH Vitamin D total level < 20 ng/ml reported as 'Deficient', 20-29 ng/ml as 'Insufficient' and 30-100 ng/ml as 'Sufficient'. The 25 OH Vitamin D TOTAL measurement sensitivity range spreads from 8.1 ng/mL up to 126.0 ng/mL. Values lower than the detectable lower limit of the measurement range of the instrument are reported as < 8.1 ng/mL and labelled as 'Severely Deficient'. The intra and inter assay repeatability and reproducibility coefficient of variation value for 25 OH vitamin D total assay were 6.4% and 10.5% for low serum range and 2.4% and 3.3% for high serum range, respectively.

Statistical analysis was performed using Windows 10 Operating system, Microsoft Office Excel 2010 and Statistical Package for Social Sciences version 20.0 (SPSS Inc, USA). Study descriptive and multiple comparison analysis was performed. p-value <0.5 was considered as statistically significant.

RESULTS

Of the 92 participants for the study, 61 of them were married (66.3%) and 31 of them were unmarried (33.7%). The mean age of the study population was 27.4± 8.2 years with range of 18-52 years. The average age of married and unmarried group was 31.1± 7.8 years and 20.2 ±1.1 years respectively and there was statistically significant difference between these groups (p=0.00). All of our participants were normotensive and consumed satisfactory Vitamin D rich food for more than 3 days within last 01 week period. None of the participants had taken Vitamin D supplementation within a month period. In our study, we found the prevalence of vitamin D deficiency was 92.4%, while that of Vitamin D insufficiency was 5.4%. Only two participants were found to have 25 OH Vitamin D total level in normal range. Both of them were married. Out of them one lady was a known case of recently diagnosed hypothyroidism and was under supplementary treatment for the diagnosis. The other lady was asymptomatic but having raised level of IgE and borderline level of impaired fasting glucose. Table 1 shows the characteristics of participants on the studied parameters with vitamin D status.

TABLE 1:CHARACTERISTICS OF VITAMIN D DEFICIENCY

Studied Parameters	Vitamin D sufficiency status(Mean±SD)				p value
	Severe VDD (n=64)	VDD (n=24)	VDI (n=5)	VDS (n=2)	
Age (years)	26.1 ±7.3	30.3±9.7	32.4±8.8	21.5± 2.1	0.058
FBS (mg/dl)	97.4± 16.3	105.3± 42.1	92.6±9.2	96±5.7	0.567
Hb% (gm/dl)	11.5±1.4	11.7±0.8	10.8±1.2	12.1± 0.4	0.542
TLC /mm3	7713.1± 2078.1	7141.7± 1411.7	7240±986.4	5550±636.4	0.277
TSH (µU/ml)	2.9±1.3	3.4±2.7	2.1±1.8	17.9±25.2	0.00
TRBC (million/ mm3)	3.9±0.3	4±0.5	3.8±0.2	4.6±0.6	0.013
Hct	34.2±3.2	34.7±2.2	33.2±2.9	36.8±1.9	0.447
MCV	88.6±7.6	87.1±8.6	87.6±4.4	79.5±5.4	0.389
MCH	29.7±3.5	29.3±3.4	28.7±2.4	26.2±3.8	0.477
MCHC	33.3±1.8	33.3±1.3	32.6±1.1	32.9±2.6	0.814
RDW	45.6±3.9	44.2±3.7	47±2.5	52.5±17.4	0.045

FBS: Fasting blood sugar; TLC : Total leucocyte count ; TSH : Thyroid stimulating hormone;TRBC : Total RBC count; Hct: Haematocrit; MCV: Mean Corpuscular volume; MCH: Mean corpuscular haemoglobin; MCHC : Mean corpuscular haemoglobin concentration; RDW: Red cell distribution width; VDD: Vitamin D deficiency;VDI: Vitamin D insufficiency; VDS : Vitamin D sufficiency

The age wise distribution shows the Vitamin D deficiency is more in the younger age groups and more so in unmarried women. (Fig 1)

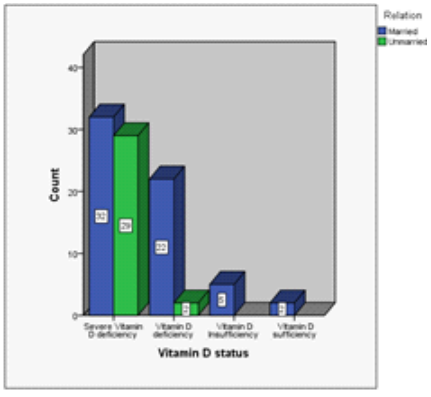
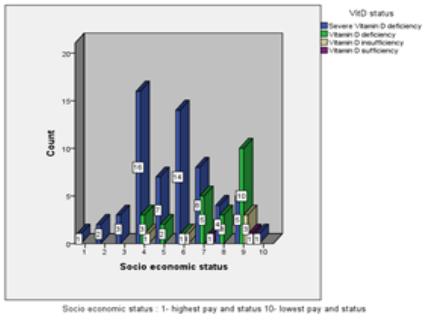


FIGURE 1: VITAMIN D LEVEL AND MARITAL STATUS

Total RBC count, RDW and TSH are the parameters which are significantly altered according to vitamin D status in the married women group. None of the studied parameters were significantly effects the vitamin D status in studied unmarried population.

There is a significant negative correlation (spearman's rho -0.420) was found with the socioeconomic status and serum vitamin D level (p=0.00). (Fig 2) The effect of socioeconomic status was significant amongst the married population (p=0.00) but did not found to effect the unmarried population.

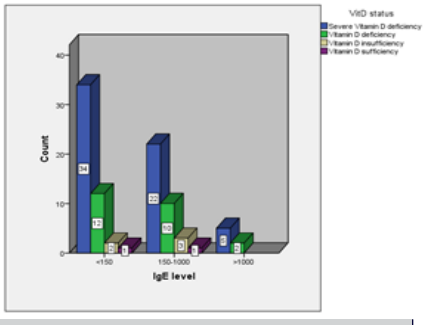
FIGURE 2: VITAMIN D STATUS AMONGST THE STUDIED SOCIOECONOMIC GROUPS



Socio economic status : 1- highest pay and status 10- lowest pay and status

Serum IgE level showed negative correlation with vitamin D level though not statistically significant. (Fig 3)

FIGURE 3: VITAMIN D STATUS AND SERUM IGE LEVEL



DISCUSSION

Our study showed the prevalence of vitamin D deficiency/ insufficiency among the studied population was 97.8%. Throughout the country, various studies reported a very high prevalence of deficiency of vitamin D even amongst healthy individuals[12]. However, one previous study from the north-eastern region on pregnant mothers by Dasgupta et al showed low prevalence (56%) of vitamin D deficiency /insufficiency[13]. No other similar comparable study is available for adult female in north-eastern region of the country. A south Indian study on reproductive female age group and north Indian study on rural adult women also showed a much lower prevalence (76.3% and 90.8% respectively) [14].

The study was conducted in the month of March in the spring season amongst educated socioeconomically well-off ladies in the urban part of north-eastern state. North eastern part of India being female-dominated society, socio-cultural tradition does not stigmatise wearing clothing that covers the majority of the skin. The study population was apparently healthy, non-hypertensive and had access to better accommodation, health care facilities and balanced diet. Therefore, the data presented most likely represent the best-case scenario; random sampling across all seasons should yield an even higher prevalence of vitamin D insufficiency. In general, Indian socio-cultural and religious culture and customs do not allow adequate sun exposure for ladies[15]. Therefore higher deficiency prevalence commensurating other parts of India may be due to the effect of culture and food habits imbibed in the migratory population.

Our study showed the occurrence of vitamin D deficiency was much more in unmarried females and became worse with the improvement of socioeconomic status. The finding is well correlated with the undying desire of unmarried Indian females to attain a fairer complexion by ways and means to avoid the sun. According to most organizations, like the World Bank and the Organization for the Economic Cooperation and Development (OECD), people with earnings in the range of US $10 to $100 per day are defined as middle class as per 2015 cost parity analysis. In our study, all the studied members studied being members of families having earnings more than Rs. 18000/month were included in the middle-class strata. It is generally observed in Indian society with the improvement of economic condition Indian women tend to adopt less outdoor exposure. Due to this peculiar social customs, our study might derive higher prevalence rate.

Even most of the educated Indians are not aware of the need of vitamin D [16]. This may be the reason even after adequate access to free health care and outdoor exposure facilities our studied population probably do not adopt preventive lifestyle changes. The traditional cooking practices in India thermally degraded vitamin D. The other important aspect of vitamin D deficiency is that it is apparently asymptomatic. In our study also we have seen there are no statistically significant differences amongst studied parameters of severely deficient and deficient unmarried ladies.

Lower 25(OH)D level was found to be associated with increased risk of hypertension [17], diabetes[18] and thyroid disorders [19]. Amongst the studied Vitamin D deficient/ insufficient population 21 ladies were having impaired fasting glucose level and 07 deficient ladies were found to have blood glucose level at a diabetic range. Altered TSH level was seen in 11 vitamin D deficient/ insufficient ladies. Vitamin D deficiency is also associated with other nutritional deficiencies. In our study we found, a significant number of vitamin D deficient participants (n=32) were having haemoglobin level below 12 gm/dl.

Vitamin D deficiency has been linked in recent studies with increased risk of immunological disorders, respiratory tract infections, asthma and allergy [20-21]. Various studies related to Vitamin D deficiencies along with a high level of IgE levels though there are some controversies[22]. It is hypothesised by experimental studies that VDR gene polymorphism along with several vitamin D associated genes, may control host utilization and/or environmental response of vitamin D which may affect serum IgE level[23]. Our study showed similar trend in a relationship between IgE and vitamin D serum level.

The main strength of the study was that this is the first study conducted of its kind in the north eastern region of the country on the adult, non-pregnant, middle class, socioeconomically stable, educated, apparently healthy, urban economically productive pan Indian population. The study investigated 25OH Vitamin D, which is the most sensitive, clinically applicable, vitamin D status parameter.

Our study has some potential limitations. The study is limited by a relatively small sample size. We have based our serum TSH, IgE and 25(OH) vitamin D thresholds based on literature provided by the kit manufacturer which is not specific for our study population. There are few inherent method specific limitations for the ELFA technique used for serum IgE, TSH and vitamin D assay. However, ELFA technique showed an acceptable coefficient of correlation (0.86) with the ideal liquid chromatography coupled to mass spectrometry method (LC-MS/MS) method. Various other previously studied important clinical and laboratory determinants of serum 25(OH)D levels, were not available for analysis. However, since important determinants of vitamin D insufficiency have been well described in this study in a well-defined identical cohort within a short span, the primary purpose of our analysis to examine the prevalence of vitamin D insufficiency in targeted population was contented.

CONCLUSIONS

This study demonstrated on the contrary to the popular belief there is an alarmingly high prevalence of vitamin D deficiency/insufficiency in the concerned population. This indicates that there is an urgent need for further effective measures required to be implemented for the spread of health awareness regarding utilisation of natural resources of vitamin D to prevent nuances of Vitamin D insufficiency/deficiency. We recommend that prevention strategies like lifestyle modification with exposure to early morning sunlight particularly for the young unmarried girls who have a high prevalence of vitamin D deficiency. Fortifying locally available staple foods with vitamin D under supplementary nutrition programmes can be a considered promotable viable population-based strategy to overcome the limitation of social barrier[24]. Vitamin D status should be considered during routine laboratory evaluation and should be followed by appropriate preventable and therapeutic measures. The finding that a large number of clinically apparently healthy population unknowingly have laboratory evidence of Vitamin D deficiency supports the need and usefulness of regular screening. Further studies are recommended on a larger cross-section of the adult urban female population to find the extent and effects of the vitamin D deficiency status and formulate a plan to eliminate this preventable health hazard.

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