Original Article

Status of Serum Vitamin D and Calcium Levels in Women of Reproductive Age in National Capital Territory of India

Nighat Yaseen Sofi[1,2], Monika Jain[1], Umesh Kapil[2], Vuthaluru Seenu[3], Lakshmy Ramakrishnan[4], Chander Prakash Yadav[5], Ravindra Mohan Pandey[6]

[1]Department of Food Science and Nutrition, Faculty of Home Science, Banasthali University, Newai, Rajasthan, Departments of [2]Human Nutrition, [3]Surgical Disciplines and [4]Cardiac Biochemistry, All India Institute of Medical Sciences, [5]National Institute of Malaria Research (NIMR), [6]Biostatistics, All India Institute of Medical Sciences, New Delhi, India

Abstract

Context: In India, Vitamin D deficiency is a major public health problem, associated with lack of sunlight exposure in spite of abundant sunshine usually accompanied by reduced dietary intake. In women of reproductive age, Vitamin D deficiency in pregnancy has been associated with an increased risk of gestational diabetes mellitus, preeclampsia, maternal and perinatal morbidity and mortality. **Aims:** The aim of the present cross-sectional study was to evaluate the levels of serum Vitamin D 25(OH) D and calcium in women of reproductive age from India. **Settings and Design:** A cross-sectional study was carried on a total of 224 healthy nonpregnant and nonlactating women in the reproductive age group of 20–49 years. **Materials and Methods:** Demographic, socioeconomic class, and biochemical parameters for the estimation of serum 25(OH)D and calcium levels in women of reproductive age were studied. **Statistical Analysis:** Statistical Package for Social Sciences version 20.0 was utilized for conducting the statistical analysis of the data. **Results:** Vitamin D deficiency (<20 ng/ml) was present in 88% of women. Women from middle socioeconomic class had the lowest mean serum 25(OH) D levels (9.6 ± 6 ng/ml) as compared to women from upper middle (11.4 ± 8 ng/ml), lower (11.2 ± 8 ng/ml), and upper (10 ± 8.6 ng/ml) socioeconomic class. Serum calcium levels were found in the normal range of 8.5–10.5 mg/dl for all the study subjects. **Conclusions:** There is a high prevalence of hypovitaminosis D among women of reproductive age. These women may possibly have a higher risk of development of osteoporosis and pregnancy-related complications in future life.

Keywords: (25(OH)D), calcium, hypovitaminosis D, reproductive age, socioeconomic class, Vitamin D

Introduction

Vitamin D deficiency is a major public health problem worldwide that has been linked not only to deficiency diseases such as rickets but to a range of common chronic diseases in adulthood including cancer, diabetes, cardiovascular health, and autoimmune diseases.[1] In women of reproductive age, Vitamin D status is of particular importance. Maternal deficiency has been associated with poor bone density, higher risk of osteoporosis particularly in women who have repeated episodes of pregnancy and lactation. Short- and long-term adverse effects in developing fetus and offspring are associated with Vitamin D deficiency.[2] Maternal Vitamin D deficiency in pregnancy has also been associated with an increased risk of preeclampsia and gestational diabetes mellitus.[3,4]

The major source of Vitamin D is sunshine exposure, even though a large percentage of the Indian population (>80%) is suffering from Vitamin D deficiency.[5] Women from reproductive age group are exposed to greater risk of developing bone abnormalities and other associated disorders due to low Vitamin D levels.[5] There is a lack of scientific literature on the status of Vitamin D and calcium in women of reproductive age from India. Hence, the present cross-sectional study was conducted to study the status of Vitamin D and calcium levels in women of reproductive age in Delhi, India.

Materials and Methods

A cross-sectional study was conducted on a total of 224 healthy women (controls) in the reproductive age group of 20–49 years.

Address for correspondence: Ms. Nighat Yaseen Sofi, Department of Food Science and Nutrition, Faculty of Home Science, Banasthali University, Newai - 304 022, Rajasthan, India.
E-mail: nighatyasinsufi@gmail.com

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The participants included were attendants of patients availing treatment from OPD services of a tertiary care hospital. The participants were included on the basis of availability over a period of 6 months. Inclusion criteria adopted was nonpregnant, nonlactating women of reproductive age without any history of chronic disease, Vitamin D intake or any other dietary supplementation and consent to participate in the study. A pretested structured questionnaire was administered to each subject to obtain information on socio-demographic profile such as name, age, educational qualification, present occupation, and monthly income of the family. The socioeconomic status of the individuals was calculated using Kuppuswamy classification.[6] Nonfasting blood samples for the biochemical from all the study individuals for serum 25(OH) D and total calcium (Ca) level estimation. Three milliliters of blood was withdrawn from the median cubital vein of individuals. The serum separation was carried out within 2 h after collection by centrifugation at 2100 rpm for 7 min. Serum samples were stored at minus 80 degree Celsius till biochemical analysis was done. Serum 25(OH) D and total calcium levels were estimated by chemiluminescent immunoassay (chemiluminescence) and colorimetric assay (Roche Cobas) technique. The values were documented in ng/ml and mg/dl. Vitamin D deficiency was defined as serum 25(OH) D levels <20 ng/ml.[7-9] The levels were further categorized under mild deficiency (10–<20 ng/ml), moderate deficiency (5–<10 ng/ml), and severe deficiency (<5 ng/ml)

Measurement of serum (25(OH)D) levels by chemiluminescence technique

Principle

25(OH) D levels in serum were measured as a standard procedure at the department of biochemistry at an apex healthcare institute. The LIAISON® 25-hydroxyvitamin D Assay (DiaSorin) uses chemiluminescent immunoassay technology. The lower limit of Quantitation of the assay was 4.0 ng/mL. (DiaSorin LIAISON® 25 OH Vitamin D TOTAL Assay [directional insert] DiaSorin, Stillwater, MN, USA; 2012) Specific antibody to Vitamin D is used for coating magnetic particles (solid phase), and Vitamin D is linked to an isoluminol derivative. During the incubation, 25(OH) D was dissociated from its binding protein and competed with labeled Vitamin D for binding sites on the antibody. After the incubation, the unbound material was removed with a wash cycle. Subsequently, the starter reagents were added, and a flash chemiluminescent reaction was initiated. The light signal was measured by a photomultiplier as relative light units and was inversely proportional to the concentration of 25(OH)D present in samples. Internal and external quality control was maintained by running a sample of known concentration of 25(OH)D along with the samples for analysis.

Measurement of serum calcium levels by colorimetric assay (Roche Cobas) technique

Serum total calcium estimation was done on an automated analyzer, COBAS INTEGRA 400 Plus. Roche Integra auto analyzer (Roche Diagnostics India Pvt. Ltd., Mumbai, India). Calcium ions react with O-cresolphthelein under alkaline conditions to form a violet colored complex. The addition of 8-hydroxylquinoline prevents interference by magnesium and ferric ions. The color intensity of the complex formed was directly proportional to the calcium concentration. It was determined by measuring the increase in absorbance at 552 nm. The value was expressed in mg/dl.

Statistical analysis

Statistical Package for Social Sciences (SPSS) version 20.0 was utilized for conducting the statistical analysis of the data (IBM SPSS statistics for Windows version 20. IBM Corp., Armonk, NY, USA). Data were expressed as mean ± standard deviation, median (lowest to highest) and frequency.

Results

The mean age of individuals was 37 ± 7 years. The percentage of individuals from nuclear and joint families was (67%, *n* = 151) and (33%, *n* = 73). The majority of the study individuals (86%, *n* = 193) were from urban areas, and (13%, *n* = 31) individuals were from rural areas. The percentages of individuals from upper socioeconomic class were (26%, *n* = 59), upper middle (33%, *n* = 73), middle (26%, *n* = 59), and lower socioeconomic class (15%, *n* = 33). The median values for serum 25(OH) D and calcium level was found to be 7.42 (4-48) ng/ml and 10 (8.5–11.8) mg/dl. It was found that 33% (*n* = 75) patients had severe Vitamin D deficiency followed by 32% (*n* = 73) patients with moderate deficiency and 22% (*n* = 49) with mild deficiency. Only 13% (*n* = 29) of subjects had serum 25(OH) D levels equal to or more than 20 ng/ml. Mean serum 25(OH) D and calcium levels for individuals from upper socioeconomic class was (10 ± 8.6 ng/ml) and (10 ± 0.5 mg/dl), upper middle (11.4 ± 8 ng/ml) and (9.8 ± 0.5 mg/dl), middle (9.6 ± 6 ng/ml) and (11 ± 0.6 mg/dl), and lower socioeconomic class (11.2 ± 8 ng/ml) and (9.8 ± 0.6 mg/dl), respectively. It was found that subjects from middle socioeconomic class had the lowest serum 25(OH) D levels as compared to subjects from upper, upper middle, and lower socioeconomic class.

Discussion

A unique property of Vitamin D is that it can be produced endogenously in the skin following sufficient sunlight exposure; specifically exposure to ultraviolet B (UVB) radiation (280–315 nm). It is regularly reported that more than 80% of Vitamin D intake is from sun exposure.[1] Other than its role in maintaining calcium and phosphorus homeostasis, promoting healthy bone mineralization, induction of cell differentiation, inhibition of cell growth, and regulation of apoptosis, Vitamin D is involved in regulating the functions of the female reproductive system. Hormones regulated by the Vitamin D system include estradiol, progesterone, human chorionic gonadotropin, and human placental lactogen, all of which are critical in maintaining the regulation of reproductive health.[10] Maternal Vitamin D deficiency or insufficiency during pregnancy has been related to preeclampsia,

gestational diabetes[11-13] conditions in bone disorder[14,15] greater risk of cesarean delivery and preterm birth.[4,14,16] It has also been associated with polycystic ovarian syndrome (PCOS), endometriosis, uterine leiomyomas, and *in vitro* fertilization outcome.[17] India is a country with ample sunshine, despite the fact, Vitamin D deficiency continues to be a growing public health concern. Other than geographic factors and ambient UVB radiation there are individual-specific variables that affect endogenous production of Vitamin D, such as limited access to sunlight, air pollution, skin condition and pigmentation (skin type), time spent outdoors, type of clothing, and sun protection practices.[18] People with darker skin require more UVB exposure (e.g., longer time outdoors) to produce Vitamin D. Exogenous sources of Vitamin D include foods and supplements. Very few foods naturally contain Vitamin D. Fatty fish, such as salmon or mackerel; contain relatively high amounts, whereas other foods, such as meats, eggs, and shellfish, contain low quantities.[19] The present study revealed that 88% of women of reproductive age group were suffering from Vitamin D deficiency (25(OH)D <20 ng/ml). The earlier studies conducted across the country have documented similar results of (96%), (76%), (58.5%), and (83%)[20-23] among women of reproductive age. Irrespective of the source, subjects from low socioeconomic status had higher mean serum (25(OH) D) levels (11.2 ± 8 ng/ml) possibly due to higher sun exposure and active lifestyle. A high prevalence of Vitamin D deficiency and poor dietary calcium intake is likely to worsen during pregnancy and may cause significant adverse health consequences in the newborn, including rickets and tetany. A recent meta-analysis concluded that Vitamin D concentrations were lower in women with PCOS compared with those without PCOS, independently of body mass index.[24] The present study reveals a high prevalence of Vitamin D deficiency among Indian women of reproductive age.

Conclusion

Vitamin D deficiency is highly prevalent across all socioeconomic groups in Indian women of reproductive age. These women may possibly have a higher risk for development of osteoporosis and pregnancy-related complications in future life. In the presence of lack of naturally occurring Vitamin D rich foods in the country, food fortification/supplementation with Vitamin D and adequate sunshine exposure should be given a higher priority among these women. There is a need of conducting more studies on a larger sample size to assess Vitamin D status in women of reproductive age group from different parts of the country to substantiate the findings of the present study.

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Conflicts of interest

There are no conflicts of interest.

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