Does diet pattern influence serum 25-hydroxyvitamin (25OHD) levels among women with polycystic ovary syndrome (PCOS): Lessons from a cross sectional study from North India?

Aafia Rashid[1*], Mohd Saleem Baba[1], Shivani Sidana[1], Mohd Afzal Zargar[2], Raman Marwaha[3], Mudasir Makdoomi[1], Namrata Sharma[4], Ishafaq Ahmed Wani[4], Mohd Ashraf Ganie[1*]

[1]Department of Endocrinology and Metabolism, Sheri-Kashmir Institute of Medical Sciences, Soura, Srinagar, J&K, India

[2]Departments of Clinical Biochemistry, University of Kashmir, India

[3]Department of Endocrinology, ILSI India, New Delhi, India

[4]Department of Gastroenterology and HNU, All India Institute of Medical Sciences, New Delhi

Abstract

Background: Polycystic ovary syndrome (PCOS), the commonest endocrinopathy of reproductive age women has multiple metabolic aberrations. VDD is rampant documented with high prevalence in India. There is a lack of substantial evidence of low VD status on the subject if vegetarians are at greater risk of VDD than non-vegetarians.

Objective: To evaluate the impact of dietary composition (veg *vs.* non-veg) on serum 25 OHD levels among women with PCOS.

Methodology: In this cross-sectional study, women aged 14–40 years who fulfilled the Rotterdam 2003 criteria for diagnosis of PCOS were enrolled from institutional clinics of AIIMS, New Delhi and SKIMS, Srinagar. Among 177 women enrolled, 74 vegetarians (from AIIMS) and 104 non-vegetarians (from SKIMS) were evaluated using a common predesigned proforma. Detailed history, clinical food frequency questionnaire (FFQ), examination and laboratory evaluation was carried out.

Results: The mean age of subjects (26.43 ± 4.44 *vs.* 26.77 ± 5.51 years) was comparable as was mean BMI (23.51 ± 2.97 *vs.* 23.81 ± 4.22 Kg/m^2).Total calorie intake was 1838.33 ± 298.54 *vs.* 1478.44 ± 404.87 kcal among non-veg and vegetarians respectively. Number of menstrual cycles/year (8.13 ± 3.40 *vs.* 8.02 ± 2.38), FG score (11.82 ± 4.52 *vs.* 11.32 ± 4.23), biochemical and hormonal parameters were also similar except for serum alkaline phosphatase (105.46 ± 32.27 *vs.* 162.70 ± 42.58 IU/L), total cholesterol (98.92 ± 24.33 *vs.* 81.15 ± 25.32 mg/dl), and triglycerides (135.01 ± 50.25 *vs.* 107.06 ± 42.10 mg/dl).The mean serum 25OHD levels did not differ significantly among non-veg and vegetarians (15.64 ± 5.41 *vs.* 12.50 ± 8.39 ng/ml).

Conclusion: Women with PCOS are linked to VDD and it is marginally severe among vegetarian women with PCOS than those consuming non-vegetarian diet.

Keywords: *PCOS, 25 OHD, Vegetarian, Total testosterone, Vitamin D deficiency.*

Accepted on July 11, 2020

Introduction

Polycystic ovary syndrome (PCOS) is the most common endocrinopathy in females of reproductive age, with the reported prevalence from 2.2% to 26% depending on which diagnostic criteria are used [1-3]. The disorder is characterized by oligo-anovulation, hyper-androgenism and/or polycystic ovarian morphology [4]. Besides, the involvement of reproductive system, metabolic disturbances are present in majority of the women suffering from PCOS. Among women with PCOS, 50 to 80% are obese, 30 to 35% have abnormal glucose tolerance (AGT), 70% have dyslipidemia, 60-70% have insulin resistance among a barrage of other metabolic constellations [5-7].

Vitamin D deficiency (VDD) is reported in epidemic proportions with a documented prevalence of 70-100% among apparently healthy Indians, similar to some published global data [8-9]. On similar lines VDD has been reported among 67-85% of women with PCOS [10-11]. VD being essential element in bone and mineral metabolism, has been linked to many non-skeletal disorders including diabetes mellitus(DM), coronary artery disease(CAD), metabolic syndrome(MS), PCOS, malignancy etc. Many studies among PCOS women report inverse relation between serum 25OHD levels and metabolic risk factors such as BMI, serum total testosterone, triglycerides, insulin resistance etc. [11-13]. This pathogenic link between vitamin D status and PCOS metabolomics is explainable on the basis of identification of VD receptor (VDR), and the 1 alpha-hydroxylase enzyme in reproductive organ tissues [14]. The finding supports this assumption that the active VD regulates over 300 genes, including genes that are important for glucose and lipid metabolism besides the gonadal function [15].

The major source of VD for most humans is casual exposure of skin to UVB (290 to 315 nm) portion of sunlight, that photolyzes 7-dehydrocholesterol (7-DHC) in the epidermis to pre-vitamin D3, followed by thermal isomerization to VD3 [16]. Although factors such as amount of solar UVB radiation, solar zenith angle, latitude, season, time of day, skin, pigmentation etc. influence endogenous synthesis of VD, many other factors (sun protection behavior, use of sunscreens, increasing interest in indoor games and indoor workplaces, concern about skin cancer etc.) prevent adequate sunlight exposure of individuals [17-18]. Under such circumstances dietary factors may become a limiting factor, which are unimportant under ordinary conditions. Sea foods (fishes like mackerel, salmon, and herring), calf liver and cod liver oil provides a good amount of VD to non-vegetarians but dairy products like milk, yoghurt, cheese and fortified cereals, juices which are low in VD content, are the only source for vegetarians [19]. This raises a question, whether vegetarians are more prone to VDD than non-vegetarians. In view of the above, this study was planned to evaluate the impact of dietary habits on the status of VD among Indian women with PCOS [20-23].

Material and Methods

In this cross-sectional study, consecutive women aged 14-40 years and diagnosed PCOS by Rotterdam 2003 criteria were enrolled from endocrinology outpatient clinics of two tertiary care clinics (All India Institute of Medical Sciences, New Delhi and Sheri-Kashmir Institute of Medical Sciences, Srinagar) located in north India. The study done from March 2014 to April 2016 was carried out in accordance with the Helsinki declaration 1975 and was approved by respective Institute Ethics Committees of AIIMS, New Delhi and SKIMS, Srinagar.

Subject selection

Clinical assessment: All consecutive women, aged 14-40 years attending these outpatient clinics for complaints of menstrual disturbances, hirsutism, acne vulgaris, androgenic alopecia, infertility etc., were informed about the study. The women who volunteered to be part of the study were asked to sign an informed consent to undergo further evaluation using a common predesigned proforma at both the centers. A detailed interview was taken about the menstrual cyclicity including age of menarche, regularity, duration, and number of cycles per year. Menstrual disturbances were classified as oligomenorrhea (≤ 8 cycles/year or menstrual interval >35 days) and amenorrhea (absence of menses in last six or more months). Other history included temporal profile of weight gain, duration of infertility, family history of PCOS, family history of type 2DM, drug intake, progression and distribution of hirsutism, severity and treatment response in acne vulgaris etc. Exclusions were made if the women were pregnant, had hyperprolactinemia, thyroid dysfunction, androgen-secreting tumours, Cushing's syndrome or Non classic congenital adrenal hyperplasia (NCAH). Women refusing consent, or with any systemic disorders or those with history of intake of drugs (VD, calcium, oral contraceptive pills, antiandrogens, insulin sensitizers or drugs known to affect glucose, insulin, or VD metabolism) in last 3 months were also excluded. A detailed diet review using a food frequency questionnaire (FFQ) and 72 hours dietary recall undertaken by qualified and trained dieticians to quantify various dietary components using specially designed diet software (Diet Cal, Profound Tech solutions, New Delhi) at both centres. For purposes of study, women who consumed meat/chicken/fish/egg at least 5 days a week at least for the preceding one year were considered as non-vegetarian and those who strictly adhered to plant and dairy based diets were taken as vegetarians.

Anthropometric assessment included measurement of height (cm), body weight (kg), waist and hip circumference (cm) with calculation of BMI (kg/m^2) using standard scales. Modified Ferriman-Gallwey (FG) score was used to assess the degree of hirsutism and a score of 8 (out of a total of 36 from nine body areas) was taken as significant [24].

Laboratory evaluation: Blood samples were collected on 3rd to 7th day (early follicular phase) of spontaneous menstrual cycle in regularly menstruating women or after medroxyprogesterone acetate withdrawal bleed. Samples were taken after an overnight (8-10h) fast with a stable diet for preceding 72 hours. The samples were aliqouted for biochemical estimations (calcium, phosphorous, liver function, kidney function, lipids) to be estimated on the same day while as aliquots for hormones (serum total T4, TSH, LH, FSH, prolactin (PRL), total testosterone, 17-OHP, cortisol (morning 8am) and 25OHD were stored at -800C for assay at a later date. The polycystic ovarian morphology (presence of 10 or more peripheral follicles each measuring 2-8mm) was assessed with trans-abdominal ultrasonography performed in the follicular phase by a single sonologist at each centre using 7.5 MHz probe (AlokaSSD-500, Tokyo, Japan) to quantitate ovarian volume, count ovarian follicle number and assess thecal hyper echogenicity.

Assays

Biochemical parameters were estimated on a fully automated biochemistry analyser (DiaSys respons®910, Germany) using standard commercially available kits with standard methodology. Hormonal (T4, TSH, LH, FSH, PRL, total testosterone, 17-OHP, cortisol assays including serum 25OHDlevels were done by electrochemiluminescence immunoassay (Cobas e411; Roche Diagnostics Limited, USA) except for plasma 17-OHP which was assayed by ELISA using commercial kits (Diagnostics Biochem, Ontario, Canada) according to supplier protocol. Glucose tolerance was categorized according to WHO 1999 criteria. A total serum testosterone >45ng/dl was taken as biochemical hyperandrogenism.

Statistical Analysis

The Statistical Package for Social Sciences 22 software was used for statistical analysis (SPSS Chicago, IL, USA). The results are represented as means ± standard deviation and log was transformed wherever necessary. Differences in concentrations of serum 25OHDlevels were evaluated by using a two-tailed unpaired Student's t test. Pearson correlation was used to correlate 25OHDlevels levels with dietary fat content. Parameters with p value <0.05 were considered significant.

Results

A total of 200 women with PCOS were enrolled from both the centres after employing inclusion criteria out of which, the data of 178 (n=74 vegetarian from AIIMS ND and n=104 non-vegetarian women from SKIMS Srinagar) was analysed after exclusions. Table 1 draws comparison of clinical, biochemical and hormonal parameters between the two groups. Briefly the groups were comparable as regards mean age (26.77 ± 5.51 *vs.* 26.43 ± 4.44 years) and mean BMI (23.51 ± 2.97 *vs.* 23.81 ± 4.22 Kg/m^2). Total calorie intake was 1838.33 ± 298.54 Kcal/day vs. 1478.44 ± 404.87 Kcal/day among non-vegetarians vs. vegetarians respectively. Non-vegetarian diets contained significantly higher amount of carbohydrates (239.86 ± 70.83 *vs.* 237.52 ± 73.26 gms), fats (39.86 ± 10.83 *vs.* 34.91 ± 15.26 gms) and proteins (55.16 ± 11.72 *vs.* 46.11 ± 13.13 gms) as compared to vegetarian diets. On further analysis, no significant difference was observed between saturated fatty acid (SFA) and monounsaturated fatty acid (MUFA) consumption between two groups, but vegetarian diets contained significantly higher amount of polyunsaturated fatty acids (PUFA). Thus, the dietary ratio of MUFA/PUFA was significantly higher in non-vegetarian diet as compared to vegetarian diet.

Number of menstrual cycles/year (8.13 ± 3.40 *vs.* 8.02 ± 2.38), FG score (11.82 ± 4.52 *vs.* 11.32 ± 4.23), biochemical (LFT, KFT, calcium, phosphorus) were also similar among non-vegetarians and vegetarians except for serum alkaline phosphatase (105.46 ± 32.27 *vs.* 162.70 ± 42.58) which was higher and total cholesterol (98.92 ± 24.33 *vs.* 81.15 ± 25.32), triglycerides (135.01 ± 50.25 *vs.* 107.06 ± 42.10) which were lower among vegetarian subgroup as compared to non-vegetarians subgroup (p<0.01). All the hormone parameters were comparable except serum25OHD (15.64 ± 15.41 *vs.* 12.50 ± 8.39 ng/ml) and total testosterone (47.42 ± 24.54 *vs.* 30.22 ± 23.31) levels, which were marginally higher among non-vegetarians.

As per IOM criteria, VDD was found between62.2% non-vegetarian and 61.2% of vegetarian women with PCOS, while 5.4% non-vegetarian and 17.4% vegetarian qualified to have insufficient VD levels (Figure 1). On the basis of cut-offs prescribed by ES guidelines, VDD and VD insufficiency were seen in 67.6% & 62.2 % of non-vegetarian and 16.25% & 28.1 % of vegetarians PCOS women (Table 2).

Table 1: Showing comparison of clinical, biochemical and hormonal parameters among vegetarian vs. non-vegetarian women with PCOS.

Characteristics	Non-vegetarian PCOS women N= 74 (Mean ± SD)	Vegetarian PCOS women N= 103 (Mean ± SD)	p value
Age (years)	26.43 ± 4.44	26.77 ± 5.51	0.646
No.of menstrual cycles /year	8.13 ± 3.40	8.02 ± 2.38	0.76
Height (cm)	157.01 ± 4.90	156.05 ± 6.07	0.24
FG score	11.82 ± 4.52	11.32 ± 4.23	0.43
Weight (Kg)	57.94 ± 7.84	57.80 ± 9.99	0.91
BMI (Kg/m^2)	23.51 ± 2.97	23.81 ± 4.22	0.58
Waist circumference (cm)	82.45 ± 9.18	83.09 ± 12.12	0.69
SBP (mmHg)	115.20 ± 8.81	116.07 ± 14.89	0.64
DBP (mmHg)	76.72 ± 8.15	76.53 ± 10.53	0.9
Serum total calcium (mg/dl)	9.24 ± 0.53	9.11 ± 0.54	0.11
Total Energy intake (kcal)	1838.33 ± 298.54	1478.44 ± 404.87	0.01
Totalcarbohydrateintake (gms/day)	304.84 ± 52.65	237.52 ± 73.26	0.01
Totalfat intake (gms/day)	39.86 ± 10.83	34.91 ± 15.26	0.01
Serum phosphorous(mg/dl)	3.60 ± 0.48	3.51 ± 0.50	0.21
Serum ALP (IU/L)	105.46 ± 32.27	162.70 ± 42.58	0.01
Fasting plasma glucose (mg/dl)	86.00 ± 9.93	87.27 ± 10.32	0.42
Serum total cholesterol (mg/dl)	167.42 ± 33.83	154.95 ± 31.51	0.01

Serum triglyceride(mg/dl)	135.01 ± 50.25	107.06 ± 42.10	0.01
Serum LDL(mg/dl)	98.92 ± 24.33	81.15 ± 25.32	0.01
Serum HDL (mg/dl)	47.50 ± 16.19	46.73 ± 11.79	0.73
Serum 25 OHD (ng/ml)	15.64 ± 5.41	12.50 ± 8.39	0.11
Serum FSH (IU/L)	6.51 ± 1.50	6.90 ± 2.67	0.26
Serum LH (IU/L)	6.98 ± 3.11	6.80 ± 2.83	0.68
Serum total testosterone (ng/ml)	47.42 ± 24.54	30.22 ± 23.31	0.01
Serum total T4 (µg/dl)	8.4 ± 1.58	8.8 ± 1.69	0.11
Serum TSH(µIU/ml)	2.79 ± 1.41	2.82 ± 1.56	0.9
Serum prolactin (ng/dl)	15.55 ± 5.62	16.07 ± 5.14	0.53

Table 2: Vitamin D status according to IOM and ES Guidelines.

Vitamin D	**IOM Guidelines**		**ES Guidelines**	
	Non-vegetarians	Vegetarian	Non-vegetarian	Vegetarian
	N (%)	N (%)	N (%)	N (%)
Deficient	46(62.2)	63(61.2)	50(67.6)	84(62.2)
Insufficient	4(5.4)	18(17.5)	12(16.2)	16(28.1)
Sufficient	24(32.4)	22(21.4)	12(16.2)	3(9.7)
Total	74(100.0)	103(100.0)	74(100.0)	103(100.0)

Note: IOM=Institute of Medicine, ES=Endocrine Society

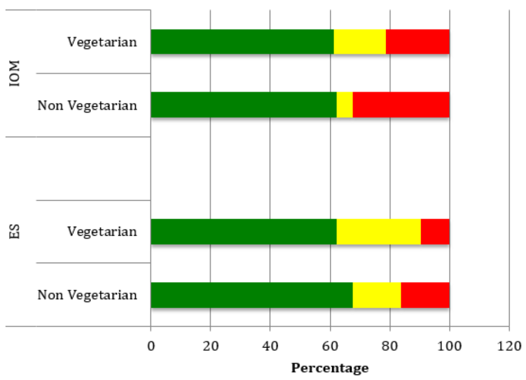

Figure 1: Showing vitamin D status among PCOS women according to different guidelines.

Discussion

VDD is linked to negative consequences on non-skeletal health including worsening of metabolic derangements among women with PCOS. Vegetarians may be at greater risk of VDD than non-vegetarians, since animal sources are believed to be rich in vitamin D. In view of the significant role played by VD in functioning of female reproductive system, the present study was intended to explore how dietary habits affect serum 25OHD levels in PCOS women and assess its impact on their metabolomics.

The main findings of the study are that PCOS women from both the centers were young (between 2^{nd} and 3^{rd} decade) and lean (BMI range 20 to 25 Kg/m^2) with matching mean age and BMI among the vegetarian and non-vegetarians subgroups. PCOS women from Srinagar were having higher mean total testosterone levels, which are consistent with our earlier findings and may be attributable to ethnic differences among these women [25]. The present study observed higher intake of calories rich in carbohydrates, fats and proteins with consequently lower serum ALP levels and elevated serum total cholesterol, LDL and TG levels among non-vegetarians as compared to vegetarians. The findings are expected and are supported by the earlier published data [26]. Higher consumption of fruit and vegetable based diets, rich in fiber, folic acid, antioxidants, and phytochemicals explain these findings. Although, dietary contents of SFA and MUFA were not different among the two groups, vegetarian diets contained significantly higher amount of PUFA and lower dietary ratio of MUFA/PUFA, no significant correlation between MUFA intake, PUFA intake or ratio of MUFA/PUFA with serum 25OHD levels was observed in both non- vegetarians and vegetarians. In the rat studies of Hollander and colleagues, it was found that higher fat in meals was associated with increased absorption of VD by increasing the solubility, changing the partition coefficient and increasing the size of the micelle [27-28]. Though, the fat content of meal did enhance absorption of the VD supplement, but the ratio of MUFA: PUFA in the meals did not influence its absorption, showing that the impact of the fat composition of meals on VD absorption and increment in serum 25OHD remains to be determined [29].

High prevalence of VDD/insufficiency as per IOM as well as ES criteria, observed both in vegetarians & non-vegetarians subgroups in the present study is consistent with the results of other studies [11]. Several factors such as poor sun exposure, sedentary life style, cultural practice of covering the body in females, use of sunscreen and increasing air pollution probably contribute to this high prevalence of VDD among Indian adolescents and adult women with PCOS. Though, the mean serum VD levels were lower in vegetarians (12.50 ± 8.39 ng/ml) as compared to non-vegetarians (15.64 ± 15.41 ng/ml), this difference was not statistically significant. VD has been increasingly recognized to play an important role in functioning of reproductive system especially of women, as it has been found to enhance key steroidogenic enzymes in granulosa cells of ovaries [30]. PCOS, the commonest endocrinopathy of reproductive age women has also been linked to VDD, besides being incriminated in worsening of metabolic parameters among PCOS women [11-12]. VDD during intrauterine life has been shown to lead to low birth weight (LBW) [31]. LBW has been associated with precocious pubarche and precocious adrenarchevia promoting insulin resistance, who are in turn at increased risk of developing PCOS [32-34]. Most of foods that are rich source of VD are generally of animal origin while unfortified milk, which is one of the main sources of VD among vegetarians, contains only 3-40 IU of VD/100ml liable to be reduced further by dilution and adulteration of milk. The only vegetarian food rich in VD is mushroom, which is not consumed regularly in adequate amounts due to lack of taste, availability and cost factor [35]. Other factors that predispose vegetarians to VDD are: vegetarian sources provide less active form of VD i.e. vitamin D2, poor absorption due to high phytate in Indian diet [36-37] and thermal degradation by deep frying [27]. In light of the above factors some studies have found higher prevalence of VDD among vegetarians [20-21]. On the contrary other hand Adventist Health Study-2, found no association between VD levels and vegetarian status [22]. Yet other studies gave contrasting results and VD levels were significantly low in non-vegetarians attributed to impaired absorption of VD by meat in the non-vegetarian diet [38].

Although the present study has several limitations such as: a) subjects were recruited from different geographical locations with different longitude, latitudes and zenith angles. b) Season of the year, skin color, duration and extent of sunlight exposure per day, pollution levels etc. were unaccounted, c) there was no control group and d) sample size was small with no PTH estimation, this is the first study among Indian women with PCOS exploring impact of dietary practices on serum 25 OH D levels.

We conclude that VDD is highly prevalent among young lean Indian women with PCOS and vegetarian diet puts them at marginally higher risk of VDD. Well-designed study on a larger cohort of subjects controlled for confounding factors is likely to address the issue.

Acknowledgement

The authors thank the study participants for their support during the study.

References

1. Chen X, Yang D, Mo Y, et al. Prevalence of polycystic ovary syndrome in unselected women from southern China. Eur J Obstet Gynecol Reprod Biol. 2008;139:59-64.
2. Azziz R, Woods KS, Reyna R, et al. The prevalence and features of the polycystic ovary syndrome in an unselected population. J Clin Endocrinol Metab. 2004;89:2745-2749.
3. Shabir I, Ganie MA, Zargar MA, et al. Prevalence of metabolic syndrome in the family members of women with polycystic ovary syndromefrom North India. Indian J Endocr Metab. 2014;18:364-369.
4. Revised 2003 consensus on diagnostic criteria and long-term health risks related to polycystic ovary syndrome The Rotterdam ESHRE/ASRM-Sponsored PCOS Consensus Workshop Group. Fertil Steril. 2004;81:19-15.
5. Dumesic DA, Oberfield SE, Stener-Victorin E, et al. Scientific Statement on the Diagnostic Criteria, Epidemiology, Pathophysiology, and Molecular Genetics of Polycystic Ovary Syndrome. Endocr Rev. 2015;36:487-525.
6. Legro RS, Kunselman AR, Dodson WC, et al. Prevalence and predictors of risk for type 2 diabetes mellitus and impaired glucose tolerance in polycystic ovary syndrome: a prospective, controlled study in 254 affected women. J Clin Endocrinol Metab. 1999;84:165-169.
7. Legro RS, Kunselman AR, Dunaif A. Prevalence and predictors of dyslipidemia in women with polycystic ovary syndrome. Am J Med. 2001;111:607-613.
8. Goswami R, Gupta A. Vitamin D deficiency in India: prevalence, causalities and interventions. Nutrients. 2014;6:729-775.
9. Hollick MF. High prevalence of vitamin D inadequacy and implications for health. Mayo Clin Proc. 2006;81:353-373.
10. Ganie MA, Marwaha RK, Nisar S, et al. Impact of hypovitaminosis D on clinical, hormonal and insulin sensitivity parameters in normal body mass index polycystic ovary syndrome women. Journal of Obstetrics and Gynaecology. 2016;36:508-512.
11. Thomson RL, Spedding S, Buckley JD. Vitamin D in the aetiology and management of polycystic ovary syndrome. Clin Endocrinol (Oxf). 2012;77:343-350.
12. Li HWR, Brereton RE, Anderson RA, et al. Vitamin D deficiency is common and associated with metabolic risk factors in patients with polycystic ovary syndrome. Metabolism. 2011;60:1475-1481.
13. Yildizhan R, Kurdoglu M, Adali E, et al. Serum 25-hydroxyvitamin D concentrations in obese and non-obese women with polycystic ovary syndrome. Arch Gynecol Obstet. 2009;280:559-563.

14. Nandi A, Sinha N, Ong E, et al. Is there a role for vitamin D in human reproduction? Horm Mol Biol Clin Investig. 2016;25:15-28.
15. Bouillon R, Carmeliet G, Verlinden L, et al. Vitamin D and human health: lessons from vitamin D receptor null mice. Endocr Rev. 2008;29:726-776.
16. Holick MF, MacLaughlin JA, Clark MB, et al. Photosynthesis of previtamin D3 in human skin and the physiologic consequences. Science. 1980;210:203-205.
17. Webb AR, Kline L, Holick MF. Influence of season and latitude on the cutaneous synthesis of vitamin D3: exposure to winter sunlight in Boston and Edmonton will not promote vitamin D3 synthesis in human skin. J Clin Endocrinol Metab. 1988;67:373-378.
18. Clemens TL, Adams JS, Henderson SL, et al. Increased skin pigment reduces the capacity of skin to synthesise vitamin D3. Lancet Lond Engl. 1982;1:74-76.
19. Misra M, Pacaud D, Petryk A, et al. Vitamin D Deficiency in Children and Its Management: Review of Current Knowledge and Recommendations. Pediatrics. 2008;122:398-417.
20. Crowe FL, Steur M, Allen NE, et al. Plasma concentrations of 25-hydroxyvitamin D in meat eaters, fish eaters, vegetarians and vegans: Results from the EPIC-Oxford study. Public Health Nutr. 2011;14:340-346.
21. Suryanarayana P, Arlappa N, Sai Santhosh V, et al. Prevalence of vitamin D deficiency and its associated factors among the urban elderly population in Hyderabad metropolitan city, South India. Ann Hum Biol. 2018;45:133-139.
22. Chan J, Jaceldo-Siegl K, Fraser GE. Serum 25-hydroxyvitamin D status of vegetarians, partial vegetarians, and nonvegetarians: the Adventist Health Study-2. Am J Clin Nutr. 2009;89:1686S-1692S.
23. Baig JA, Sheikh SA, Islam I, et al. Vitamin D status among vegetarians and non-vegetarians. J Ayub Med Coll Abbottabad JAMC. 2013;25:152-155.
24. Aswini R, Jayapalan S. Modified Ferriman-Gallwey Score in Hirsutism and its Association with Metabolic Syndrome. Int J Trichology. 2017;9:7-13.
25. Ganie MA, Marwaha RK, Dhingra A, et al. Observation of phenotypic variation among Indian women with polycystic ovary syndrome (PCOS) from Delhi and Srinagar. Gynecol Endocrinol. 2016;32:566-570.
26. Kumar Das S, Faruque ASG. Comparative Study of Lipid Profile Levels in Vegetarian and Non-Vegetarian Person.IJLSSR. 2015;1:89-93.
27. Hollander D, Muralidhara KS, Zimmerman A. Vitamin D-3 intestinal absorption in vivo: influence of fatty acids, bile salts, and perfusate pH on absorption. Gut. 1978;19:267-272.
28. Hollander D. Intestinal absorption of vitamins A, E, D, and K. J Lab Clin Med. 1981;97:449-462.
29. Dawson-Hughes B, Harris SS, Lichtenstein AH, et al. Dietary fat increases vitamin D-3 absorption. J Acad Nutr Diet. 2015;115:225-230.
30. Merhi Z, Doswell A, Krebs K, et al. Vitamin D Alters Genes Involved in Follicular Development and Steroidogenesis in Human Cumulus Granulosa Cells. J Clin Endocrinol Metab. 2014;99:E1137-1145.
31. Burris HH, Rifas-Shiman SL, Camargo CA, et al. Plasma 25-Hydroxyvitamin D During Pregnancy & Small-for-Gestational Age in Black and White Infants. Ann Epidemiol. 2012;22:581-586.
32. Ibáñez L, Potau N, Marcos MV, et al. Exaggerated adrenarche and hyperinsulinism in adolescent girls born small for gestational age. J Clin Endocrinol Metab. 1999;84:4739-4741.
33. Ibáñez L, Dimartino-Nardi J, Potau N, et al. Premature adrenarche--normal variant or forerunner of adult disease? Endocr Rev. 2000;21:671-696.
34. Wehr E, Pilz S, Boehm BO, et al. Association of vitamin D status with serum androgen levels in men. Clin Endocrinol (Oxf). 2010;73:243-248.
35. Ko JA, Lee BH, Lee JS, et al. Effect of UV-B exposure on the concentration of vitamin D2 in sliced shiitake mushroom (Lentinus edodes) and white button mushroom (Agaricus bisporus). J Agric Food Chem. 2008;56:3671-3674.
36. Heaney RP, Recker RR, Grote J, et al. Vitamin D(3) is more potent than vitamin D(2) in humans. J Clin Endocrinol Metab. 2011;96:E447-452.
37. Harinarayan CV, Ramalakshmi T, Prasad UV, et al. High prevalence of low dietary calcium, high phytate consumption, and vitamin D deficiency in healthy south Indians. Am J Clin Nutr. 2007;85:1062-1067.
38. Mawer EB, Davies M. Vitamin D nutrition and bone disease in adults. Rev Endocr Metab Disord. 2001;2:153-164.

***Correspondence to**

Mohd Ashraf Ganie,

Department of Endocrinology and Metabolism,

Sheri-Kashmir Institute of Medical Sciences Srinagar, India

Phone: +91-194-2405501

Email: ashraf.endo@gmail.com