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Original Research Article

Frequency of Hypovitaminosis D in Obese Adolescent with Polycystic Ovary Syndrome

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Abstract

***Background*:** Polycystic ovary syndrome (PCOS) is one of the most common female endocrine disorders affecting the reproductive age group and is thought to be one of the leading cause of female infertility. Many patients with polycystic ovary syndrome (PCOS) also have features of the metabolic syndrome, including insulin resistance, obesity, and dyslipidemia. Vitamin D affects insulin and glucose metabolism, and a low vitamin D status maybe a risk factor for PCOS. ***Aim*:** To determine vitamin D status among the obese adolescent with Polycystic Ovary Syndrome (PCOS) and to compare the vitamin D level among obese and nonobese adolescent with PCOS. ***Methods*:** This cross sectional study was done at the out-patient department of Obstetrics and Gynecology in Bangabandhu Sheikh Mujib Medical University (BSMMU) during November 2017 to October 2018 but the patients enrollment was started after 16 July 2018. The study comprises of 80 adolescent girls diagnosed as PCOS according to Rotterdam criteria, in the age range of 9 to 18 years, out of which 50 obese adolescent girls was considered as group I and 30 non obese adolescent girls was considered as group II. The biochemical parameters measured in the study includes fasting blood sugar, fasting insulin level and serum 25-hydroxy vitamin D level. Serum 25 (OH) D level less than 20 ng/ml was classified as vitamin D deficiency. ***Results*:** In this study it was observed that maximum patients were vitamin D deficient in both groups. In group I, 76% and in group II, 60% were vitamin D deficient. Again in group I, 14% and 33.3% in group II were vitamin D insufficient. Only 10% in group I and 6.7% in group II were found to have sufficient level of vitamin D. The mean Vitamin D was found 18.25±5.51 ng/ml in group I and 19.66±5.21 ng/ml in group II. The mean difference of vitamin D level between two groups was statistically not significant. There is a negative significant correlation between vitamin D and BMI which indicates level of vitamin D decreases with increase of BMI of the participants. ***Conclusion*:** Vitamin D deficiency was predominant in both groups. Though the difference of vitamin D level was not significant between two groups but the mean level of vitamin D is lower in group I than in group II. There is a negative significant correlation between vitamin D and BMI which indicates that higher BMI levels were associated with lower vitamin D levels.

Keywords: Hypovitaminosis, Polycystic Ovary Syndrome, Vitamin D.

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INTRODUCTION

Polycystic ovary syndrome (PCOS) is a very common endocrine disorder which is present in approximately 7% of reproductive age women [1]. It is a heterogeneous syndrome that usually presents during adolescence and is characterized by features of anovulation (amenorrhea, oligomenorrhea, irregular

menstrual cycles) combined with symptoms of androgen excess (hirsutism, acne, alopecia) and insulin resistance. As the symptoms of PCOS begin usually in adolescence but, it is believed that its origin lies in childhood or fetal life, though, it is not diagnosed until 2-3 years after menarche [2]. It is suggested that there is a link between vitamin-D and the pathogenesis of obesity in adults and children [3]. Vitamin D is thought to influence the development of PCOS through gene transcription [4]. Hormonal modulation influences insulin metabolism and fertility regulation [5]. Vitamin D plays an essential role in regulating body levels of calcium and phosphorus. Bone growth and maintenance of bone remodeling mostly depends on vitamin D, [6] which are particularly important processes in childhood and adolescence. However, in recent years link between vitamin deficiency and the pathogenesis of several chronic conditions such as metabolic syndrome, type 2 diabetes, hypertension, obesity, increased risk of cardiovascular disease, autoimmune thyroid diseases and development of PCOS has been found [7]. The scientific literature has reported high prevalence of hypovitaminosis D in the world, not only in vulnerable individuals, such as older adults and postmenopausal women, [8] but also among the children and adolescents [9]. Several studies have shown that low serum 25(OH) D levels are associated with high body mass index (BMI), insulin resistance and waist / hip circumference, but it has not yet been clearly determined [10]. Binding of vitamin D and vitamin D receptors may increase the transcriptional activation of the insulin gene and the synthesis of insulin. It also stimulates the activation of glucose transporters. On the other hand, one of the functions of vitamin D is to regulate the calcium concentration in B cells. Calcium is also essential for the insulin-mediated intracellular process in insulin target tissues. The factors that have been associated with lower levels of serum calcidiol are gender, age, race, sun exposure season of the year in which serum is collected, and childhood obesity [11]. Vitamin D deficiency in obese individuals is attributed to several factors, such as decreased exposure to sunlight due to sedentary lifestyle, or to excessive vitamin sequestration within adipose tissue [12]. Furthermore, significant evidences also relates this phenomenon to behavioral, dietary and genetic factors, habits such as excessive sunscreen use, low consumption of dietary sources of vitamin D, or most importantly the genetic predisposition of individuals could contribute to the increase of hypovitaminosis D [13]. As it is unknown whether obese adolescent girls with PCOS have higher prevalence of vitamin D deficiency as compare to nonobese adolescent girls with PCOS, the aim of our study was to compare the serum 25 (OH) D level in PCOS subjects according to obesity.

MATERIALS AND METHODS

Place of study: Department of Obstetrics and Gynaecology of BSMMU, Dhaka, Bangladesh.

Period of study: 1 year (November 17 to October 18) but the patients enrollment was started after IRB clearance (16 July 2018).

Study design: Cross-sectional study.

Study population: A total of 80 adolescent girls aged 9 to 18 years having PCOS (which was diagnosed according to Rotterdam criteria) were selected from the OPD of Obstetrics and Gynaecology and as well as OPD of endocrinology of BSMMU. Among them 50 were obese adolescent and 30 were nonobese adolescent.

Sample Size:

To determine the sample size the following formula was followed

$$n=\frac{(Za+ZB)^2\ x\ (0_1{}^2+0_2{}^2)}{(\mu_1-\mu_2)^2}=n=30$$

Here, the values of mean and SD of two groups were taken from the study [14]. According to this formula though the targeted sample was 30. But to increase the power of the study more sample as much as possible was taken. Finally total 80 patients, 50 obese as group I and 30 nonobese as group II were included.

Inclusion Criteria:

1. PCOS was diagnosed according to the Rotterdam criteria: The presence of the following any two of these three criteria
   - I. Menstrual abnormalities like amenorrhoea (no cycles in the past 6 months), oligomenorrhoea (cycles lasting longer than 35 days), or long cycles
   - II. Clinical and/or biochemical hyperandrogenism, (hirsutism, acne, alopecia etc)
   - III. Ultrasound (USG) appearance of polycystic ovaries (multiple cysts >12 in number and 2-9 mm in size).
2. Adolescent girls age belonged to 9 - 18 years.
3. Obese patient (Obesity is defined as a BMI at or above the 95th percentile for children and teens of the same age and sex).

Exclusion Criteria: Exclusion based on clinical assessment and previously diagnosed cases:

- Congenital adrenal hyperplasia
- Cushing's syndrome
- Adrenal tumour
- Ovarian tumour
- Thyroid dysfunction
- Prolactinoma or hyperprolactinemia
- Patient who are taking vitamin D supplement
- PCOS patient who are taking Metformin, OCP, Lipid lowering agent
- Patient who refused to participate in the study

Study Procedure:

This prospective study was conducted in Bangabandhu Sheikh Mujib Medical University

(BSMMU), Dhaka, Bangladesh. A total of 80 adolescent girls of 9-18 years of age with PCOS attending the OPD of Obstetrics and Gynaecology and OPD of endocrinology, BSMMU were recruited for the study. After fulfilling the inclusion and excluding the exclusion criteria, patients were enrolled with unique ID. Subjects were briefed about the objectives of the study, risk and benefits for participating in the study and confidentiality. Patients were allocated into two groups, of which 50 were obese adolescent as group I and 30 were nonobese adolescent as group II. Written informed consent was obtained form each patients and from their guardians as well after careful explanation of the study procedure. Detailed history was taken about their educational status, area of residence, monthly family income. Regarding personal history exposure to sunlight, during sun exposure average surface area of the body covered with cloths or not, use of sunscreen lotion were asked for. To find out dietary intake of vitamin D patients were asked about consumption of milk, egg and small or oily fish. The weight and height of all participants were measured. The patients were weighed using spring balance with minimum clothing after correcting zero error. The weight was recorded to the nearest 500 gm. The height was measured keeping the women standing on level ground, without footwear, against a wall, by using measuring tape to the nearest of 1 cm. Patients weight and height was obtained to calculate BMI (kg/ m^2). According to operational definition obesity was defined as a BMI at or above the 95th percentile for children and teens of the same age and sex and overweight was defined as a BMI at or above the 85th percentile and below the 95th percentile for children and teens of the same age and sex. Patient's waist circumference and hip circumference was also measured and recorded. Blood sample was collected for measurement of 25 (OH) D level, fasting blood sugar and fasting insulin level.

Preparing and Organizing Blood Sample:

Blood samples were collected from department of Obstetrics and Gynaecology of Bangabandhu Sheikh Mujib Medical University (BSMMU) after taking informed written consent from subjects who fulfill the inclusion criteria. Data was collected in pretested semi-structured questionnaire after completion of detail history and physical examination. Venous blood was collected by venipuncture with the subject sitting comfortably in a chair in a quiet room. Blood collected test tube immediately covered by aluminum foil paper. After 10-15 minutes of collection, blood sample was centrifuged for 10-15 minutes at 3000 rpm in a dark room to obtain serum and it send to the laboratory without delay. Sample was preserved appropriately and were be analyzed for serum 25(OH) D within a week of sample collection.

Assay of serum 25(OH) vitamin D:

Assay of serum 25(OH) D of collected sample was done in Centre for Advanced Research in Sciences (CARS), University of Dhaka by competent laboratory personal. Vitamin D was measured using automated analyzer by High Performance Liquid Chromatography (HPLC) for the quantitative determination of 25(OH) D in human plasma by HPLC 25-OH-D assay.

Principles of the test: [15]

HPLC is a technique in analytical chemistry used to separate, identify, and quantify each component in a mixture. It relies on pumps to pass a pressurized liquid solvent containing the sample mixture through a column filled with a solid adsorbent_material. The components of the sample mixture are separated from each other due to their different degrees of interaction with the adsorbent particles. The detector generates a signal proportional to the amount of sample component emerging from the column, hence allowing for quantitative analysis of the sample components. A digital microprocessor and user software control the HPLC instrument and provide data analysis.

Procedure of the test:

0.5 ml of blood sample was added with 350 µl of methanol and 2-propanol in the ratio of 80:20 (v/v). The contents were mixed in a vortex mixer for 30s. Vitamin D was extracted by mixing two times (60 s each time) with 1 ml of hexane. The phases were separated by centrifugation, and, the upper organic phase was transferred to a conical tube and dried under nitrogen. The residue was dissolved in appropriate volume of mobile phase. The mobile phase was filtered through a 0.45 um membrane filter and passed through column at 0.4 ml min^1 for column equilibration; the baseline was monitored continuously during this process. Detection was carried out at λ max 265 nm. The prepared dilutions were injected in series, peak area was calculated for each dilution, and concentration was plotted against peak area. The method described here is designed to detect serum 25(OH) D values from 5 to 100 ng/ml.

Statistical Analysis of Data:

All the relevant collected data was compiled on a master chart first. Then organized by using scientific calculator and standard statistical formula. Percentages were calculated to find out the proportion of the findings. Further statistical analysis of the results was done by Microsoft excel. The results were presented in tables and figures. Statistical analysis of BMI and vitamin D were done by unpaired t-test. Statistical analysis of relationship between vitamin D and BMI was done by Pearson correlation. P-value<0.05 was considered statistically significant.

RESULTS

Table 1: Distribution of the study population by demographic variables (n=80)

Demographic variables	Group-I (n=50)		Group-II (n=30)	
	Mean±SD		Mean±SD	
Age (in year)	14.9±2.36		15.93±1.72	
Range	9-18		12-18	
Educational status	**n**	**%**	**n**	**%**
Primary	2	4.0	0	0.0
Secondary	34	68.0	18	60.0
Higher Secondary	14	28.0	12	40.0
Area of residence				
Urban	42	84.0	30	100.0
Rural	8	16.0	0	0.0
Monthly family income (in thousand taka)				
<30	2	4.0	0	0.0
30-60	20	40.0	12	40.0
>60	28	56.0	18	60.0

Group-I= Obese, Group-II Nonobese

Table 1 showing the demographic variable of the study population. The mean age was found 14.9±2.36 years in group I and 15.93±1.72 years in group II. More than two third (68.8%) patients were in their secondary education level in group I and 18 (60.0%) in group II. In group I, maximum patients came from urban area and in group II all patients were from urban area. Maximum patients of both groups belonged to family income >60000 taka per month which was 24 (48.0%) in group I and 18 (60.0%) in group II. Family income was categorized according to new country classifications by income level Submitted by World Bank Data Team.

Table 2: Distribution of the study population by personal history (n=80)

Personal history	Group-I (n=50)		Group-II (n=30)	
	n	%	n	%
Sunlight exposure (in hour /day)				
<1 hour	3	6.0	4	13.3
1 hour	24	48.0	10	33.3
>1 hour	23	46.0	16	53.3
Average surface area of the body covered with cloths				
Yes	50	100.0	30	100.0
No	0	0.0	0	0.0
Use of sunscreen lotion during sun exposure				
Yes	4	8.0	0	0.0
No	46	92.0	30	100.0
Physical activities (Daily exercise or participation in sports)				
Yes	15	30.0	8	26.7
No	35	70.0	22	73.3

Table 2 showing the personal history of the study population. About sunlight exposure maximum patients gave history of exposure in an average 1 hour per day in both groups, which was 24 (48.0%) in group I and 10 (33.3%) in group II. In both groups all patients 50 (100%) in group I and 30 (100%) in group II had their average surface area of the body covered with cloths and not exposed to sunlight. During sun exposure only 4 (8.0%) patients used sunscreen lotion in group I and no patient in group II. 15 (30.0%) in group I and 8 (26.7%) in group II gave history of daily exercise or participation in sports.

Table 3: Distribution of the study population by usual food habit (n=80)

	Group-I (n=50)		Group-II (n=30)	
	n	%	n	%
Milk consumption 4 times a week				
Yes	32	64.0	22	73.3
No	18	36.0	8	26.7
Egg consumption per day				
Yes	41	82.0	28	93.3
No	9	18.0	2	6.7
Small fish intake once a week				
Yes	19	38.0	12	40.0
No	31	62.0	18	60.0
Fast food consumption >2times a week				
Yes	20	40.0	8	26.7
No	30	60.0	22	73.3

Table 3 showing the usual food habit of the study population. Maximum patients had milk consumption 4 times a week in both groups, which was 32 (64.0%) in I and 22 (73.3%) in group II. About consumption of egg per day in group I, maximum patients take 41 (82.0%) and 28 (93.3%) in group II. Minimum patients took small fish once a week in both groups, which was 19 (38.0%) in group I and 12 (40.0%) in group II. 20 (40.0%) in group I and 8 (26.7%) in group II gave history of intake of fast food more than two times a week.

Table 4: Distribution of clinical findings of the study population (n=80)

Clinical Examination	Group-I (n=50)	Group-II (n=30)	p value
	Mean±SD	Mean±SD	
Weight (in kg)	79.12±12.46	55.87±7.3	0.001[s]
Range	49-105	45-73	
Height (in cm)	157.14± 6.4	160.53±2.29	0.007[s]
Range	139-170	158-165	
BMI (kg/m²)	31.99±4.51	21.66 ±2.82	0.001[s]
Range	25.1-39.7	18.0-24.8	
Waist/ Hip Ratio	0.88± 0.03	0.82±0.04	0.001[s]
Range	0.84-0.92	0.75 0.87	

s=significant, p value reached from unpaired t-test

Table 4 showing the Clinical Examination of the study population. The mean weight was found 79.12±12.46 kg in group I and 55.87±7.3 kg in group II. The mean height was found 157.14±6.4 cm in group I and 160.53±2.29 cm in group II. The mean BMI was found 31.99±4.51 kg/m² in group I and 21.66±2.82 kg/m² in group II. The mean of waist / hip ration in group I was 0.88± 0.03 in group I and 0.82±0.04 in group II. The difference was statistically significant (P<0.05) between two groups.

Table 5: Laboratory parameters of the study populations (n=80)

Laboratory parameters	Group-I (n=50)	Group-II (n=30)
	Mean±SD	Mean±SD
Fasting blood sugar (mg/dl)	5.07±0.58	4.59±0.95
Range	3.9-6.3	1.5-5.5
Fasting insulin (µU/ml)	18.7 ±7.72	16.79±7.16
Range	4.3-32.06	4.23-30.7

Table 5 showing the Laboratory parameters of the study population. The mean fasting blood sugar was found 5.07±0.58 mg/dl in group I and 4.59±0.95 mg/dl in group II. The mean fasting insulin was found 18.7±7.72 µU/ml in group I and 16.79±7.16 HU/ml in group II.

Table 6: Comparison between vitamin D with BMI (n=80)

BMI (kg/m²)	Vitamin D (ng/ml)		p value
	Group-I (n=50)	Group-II (n=30)	
	Mean±SD	Mean±SD	
18-24	24.56±0.71	19.80±5.37	0.001[s]
25-30	19.56±6.03	17.12±0.73	0.031[s]
>30	17.03±4.94		

s=significant, p value reached from unpaired t-test

Table 6 showing the mean vitamin D level was found 24.56±0.71 ng/ml belonged to BMI 18-24 kg/m² in group I and 19.80±5.37 ng/ml in group II. The mean vitamin D level was found 19.56±6.03 ng/ml belonging to BMI 25-30 kg/m²in group I and 17.12±0.73 ng/ml in group II. The mean vitamin D was found 17.03±4.94 ng/ml belong to BMI >30 kg/m² in group I and not found in group II. The difference was statistically significant (P<0.05) between two groups.

Table 7: Vitamin D status of the study population (n=80)

Vitamin D (ng/ml)	Group-I (n=50)		Group-II (n=30)		p value
	n	%	n	%	
Deficient (20)	38	76.0	18	60.0	
Insufficient (21-29)	7	14.0	10	33.3	
Sufficient (230)	5	10.0	2	6.7	
Mean+SD	18.25±5.51		19.66±5.21		[a]0.261[ns]
Range	9.74-30.92		11.16-30.96		

Ns=not significant, p value reached from unpaired t-test

Table 7 showing the vitamin D status of the study population. Maximum patients were vitamin D deficient that is ≤20 ng/ml in both groups which was 38 (76.0%) in group I and 18 (60.0 %) in group II. The mean Vitamin D level was found 18.25±5.51 ng/ml in group I and 19.66±15.21 ng/ml in group II. The difference was statistically not significant (P<0.05) between two groups.

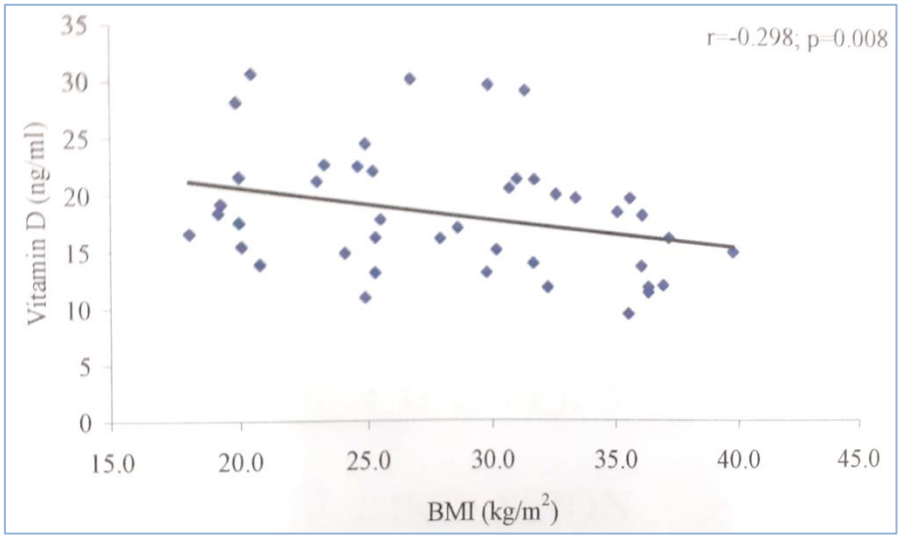

Figure 1: Scatter diagram showing Pearson's correlation between vitamin D and BMI.

Figure 1 showing negative significance of Pearson's correlation (r= 0.298; p=0.008) between vitamin D and BMI.

DISCUSSION

PCOS is a multifactorial disorder, several studies have been performed to determine this phenomena. He role of vitamin D in PCOS, and its possible implication for metabolc disorders among the PCOS patients, has been studied in recent years. Low vitamin D status is suspected to be a risk factor for insulin resistance and obesity on the other hand obesity may have role in vitamin D deficiency. This current cross sectional study was carried out with an aim to measure

vitamin D level in obese and nonobese adolescent with PCOS and to categories them accordingly. Comparison of Vitamin D level was done between obese and nonobese adolescent with PCOS. A total of 80 adolescent fulfilling the Rotterdam criteria for diagnosis of PCOS attending in the out-patient department of Obstetrics and gynecology and department of endocrinology and metabolism in Bangabandhu Sheikh Mujib Medical University (BSMMU), Dhaka, were included in this study. Among them 50 obese adolescent were considered as group I and 30 nonobese adolescent were considered as group II. Adolescent girls who are age belonged to 9 - 18 years were enrolled in this study. Congenital adrenal hyperplasia, cushing's syndrome, adrenal tumour, ovarian tumour, thyroid dysfunction, prolactinoma or hyperprolactinemia patient who are taking vitamin D supplement, PCOS patient who are taking Metformin, OCP, Lipid lowering agent and patient who refused to participate in the study were excluded from the study. In this current study, it was observed that maximum patients had Vitamin D deficiency that is 25 (OH) D level 20 ng/ml in group I was 76.0% and 60.0% in group II. 14% and 33.3% patients were vitamin D insufficient in group I and group II group I respectively. Only 10% and 6.7% patient had normal level of vitamin D in and group II respectively. The mean Vitamin D was found 18.25±5.51 ng/ml in group I and 19.66±5.21 ng/ml in group II. The difference was statistically not significant (P<0.05) between two groups. Bostanci et al., [16] observed a high prevalence of vitamin D deficiency in both PCOS and control group subjects. In another study Sadhir et al., [17] observed Vitamin D deficiency was noted among more than the half of the participants with PCOS and also vitamin D deficiency was found in almost half of participants of the control group. In this present study, it was observed that the mean age was found 14.9±2.36 years in group I and 15.93±1.72 years in group II. Sadhir et al., [17] observed that the mean in their study sample was 15.2 years, which support with the present study. In this current study, it was observed that the 68.0% patients were in their secondary education level in group I and 60.0% in group II and patients in their higher secondary level were found 28.0% and 40.0% in group I and group II respectively. Primary level found only 4.0% in group I but not found in group II. Similarly, Shawna et al., [18] conducted their study among them high school patients with PCOS and in non-PCOS. Regarding the area of residence, it was observed that 84.0% patients came from urban area in group I and 100.0% in group II. Dura-Trave et al., [19] and Williams et al., [20] studies observed that most of the patients are from urban residence, which are similar with the present study. In this current study, it was observed that maximum patients of both groups belonged to family income >60000 taka per month in both groups, which was 48.0% in group I and 60.0% in group II. On the other hand, Chauhan et al., [21] observed that majority of the adolescents came from lower socioeconomic strata in this current study, it was observed that among history of sunlight exposure maximum patients gave history of sunlight exposure about 1 hour per day in both groups, which was 48.0% in group I and 33.3% in group II. Though, in this present study, it was observed that 100.0% patients had most of the surface area of their body covered with cloths in both groups. Regarding the usual food habit, it was observed in this current study that maximum patients had taken milk age and small fish in both groups but the patient had low level of vitamin D. Regarding the fast food consumption >2times a week it was observed that 40.0% in group I and 26.7% in group II took that. Williams et al., [20] mentioned that the etiology of hypovitaminosis D is likely multifactorial because it has been associated with several dietary factors as well as decreased sunlight exposure poor vitamin D intake. In this current study, it was observed that the mean weight was found 79.12±12.46 kg in group I and 55.87±7.3 kg in group II. Similarly, the mean height was found 157.14±6.4 cm in group I and 160.53±2.29 cm in group II. The mean BMI was found 31.99±4.51 kg/m² in group I and 21.66±2.82 kg/m² in group II. The mean of waist / hip ratio in group I was 0.88±0.03 and 0.82±0.04 in group II. The difference was statistically significantly (P<0.05) higher in group I. Shawna et al., [18] obtained in their study that excessive body weight was significantly associated with PCOS. Coviello et al., [22] reported an association between PCOS and metabolic syndrome independent of body mass index (BMI); obese adolescents with PCOS had a greater prevalence of metabolic syndrome than obese adolescents without PCOS. Regarding the Fasting blood sugar, it was observed in this present study that the mean fasting blood sugar was found 5.07±0.58 mg/dl in group I and 4.59±0.95 mg/dl in group II, which was significantly higher in group I. In this current study, it was observed that the mean fasting insulin was found 18.7±7.72 µU/ml in group I and 16.79±7.16 µU/ml in group II. In this study there was a negative significant correlation (r=-0.298; p=0.008) found between vitamin D (ng/ml) and BMI (kg/m²), which indicates that higher BMI levels were associated with lower vitamin D levels. Bostanci et al., [16] found a high prevalence of vitamin D deficiency in both PCOS and control group. Although the serum 25(OH) D levels were lower in the PCOS group when compared to the non- PCOS group, there were no statistically significant differences among them. Li et al., [5] indicated no significant difference in serum 25(OH) D levels between patients and controls. Mahmoudi et al., [4] reported quite higher levels of serum 25(OH) D in PCOS patients compared to the control group with a similar BMI and age. In present study there are several limitations that should be noted. Firstly the sample size was small and not equal in both groups. The study was conducted in tertiary care centre so over all status of the whole community was not reflected. The study did not find any relationship between serum 25(OH) D levels and clinical and metabolic profiles in two groups.

CONCLUSION

This study was undertaken to determine vitamin D status among the obese adolescent with Polycystic Ovary Syndrome (PCOS). Vitamin D deficiency was predominant in both groups. Though the difference of vitamin D level was not significant between two groups but the mean level of vitamin D is lower in group I than in group II. There is a negative significant correlation between vitamin D and BMI which indicates that higher BMI levels were associated with lower vitamin D levels.

Limitations

1. The study population was selected from one selected hospital in Dhaka city, so that the results of the study may not be reflect the exact status of the country.
2. The present study was conducted at a very short period of time. So association of vitamin D deficiency with seasonal variation was not found.
3. This study did not find any relationship between serum 25(OH) D levels and clinical or metabolic profiles in two groups.
4. In addition, sample size in each group was smaller and not equal in both groups.
5. Samples were taken by purposive method in which question of personal biasness might arise.

RECOMMENDATIONS

Obesity could be considered as an associated factor for vitamin D deficiency of PCOS patients and, owing to its high prevalence, the implementation of systematic screening and hypovitaminosis D treatment programs would be particularly useful. Multicentric studies with large sample size may be carried out in future. Further studies in adolescent females with PCOS and normal BMI could be helpful in delineating the role of Vitamin D in pathogenesis of PCOS. Studies can be done after vitamin D supplementation in obese patient with or without PCOS and outcome can be observed.

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