

A Comparative Study on the Role of Vitamin D3 supplementation among Poly Cystic Ovarian Syndrome Women with Vitamin D3 deficiency**Kancharla Venkata Srilakshmi Devi^{1*}, Konide Sai Anudeep², B.Venkateswara Rao³**¹Associate Professor, Department of Obstetrics & Gynaecology, Katuri Medical College & Hospital, Chinnakondrupadu, Guntur, Andhra Pradesh, India²Internee of Katuri Medical College, Chinnakondrupadu, Guntur, Andhra Pradesh, India³Professor and HOD, Department of Obstetrics & Gynaecology, Katuri Medical College & Hospital, Chinnakondrupadu, Guntur, Andhra Pradesh, India

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Conflict of interest: Nil

Abstract:**Aim:** To determine the effectiveness of Vitamin-D3 supplementation with Metformin in woman with established diagnosis of PCOS & Vitamin D3 deficiency.**Method:** A total of 100 women diagnosed with PCOS with Vitamin D3 deficiency fulfilling the inclusion criteria were included in to the study and are sub divided in to two groups A & B with 50 women in each group. Group A Women were given treatment with Metformin & Calcium 1000mg daily along with Vitamin D3 60K IU weekly for 6weeks followed by monthly for 4 months. Group B Women were given treatment with Metformin 1000mg daily only.**Results:** On comparing between the two groups, it was found that addition of Vitamin D & Calcium to Metformin regularize the menstrual cycles significantly with p value 0.031. In relation to mean ovarian volume, after 6months of treatment there was a significant reduction by 30% (p <0.0001) in group A i.e Vitamin D, Calcium & Metformin supplemented women. Regarding BMI, waist hip ratio, acne, hirsutism & serum fasting insulin there was reduction in mean values after treatment for 6months in both groups, but reduction is more in group A (Vitamin D, Calcium & Metformin) than in group B women (Metformin). But this difference was not statistically significant.**Conclusion:** The Vitamin D3 & Calcium are safe, inexpensive & easily available drugs their addition to Metformin can contribute for normalization of the dysregulated metabolism in various tissues including ovaries, pancreas, and muscle and enhance the action of Metformin in improving the clinical biochemical features of PCOS.**Keywords:** Polycystic ovary syndrome,

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Introduction

Polycystic ovary syndrome (PCOS) is the most common endocrine disorder of women of reproductive age affecting about 6-15% of all women. A complex interaction of various factors contributes to the pathogenesis of PCOS. The most widely accepted theory proposes that PCOS is a self-perpetuating cycle of hormonal events with arrested follicular development, increased androgen concentration resulting in polycystic ovaries. [1-5]

PCOS is characterized by increased ovarian and adrenal androgen secretion, hyperandrogenic symptoms like hirsutism, acne, alopecia, menstrual irregularities & polycystic ovaries with increased stromal thickness & ovarian volume. [6] PCOS is associated with multiple risk factors such as insulin resistance, central obesity, subclinical atherosclerosis, hypertension, impaired glucose tolerance,

type2 DM, metabolic syndrome, dyslipidaemia, infertility, endometrial hyperplasia, endometrial & ovarian cancers. [3] Early detection and treatment of the disease is absolutely important. Diagnosing PCOS is not easy as signs and symptoms are heterogeneous and vary from time to time. Diagnostic criteria have been summarized by the European Society for Human Reproduction (ESHRE) and the American Society for Reproductive Medicine (ASRM) in 2003 and denominated as 'Rotterdam Criteria'. [7] In 2006 Androgen excess society developed a criteria making androgen excess as a necessary component for diagnosing PCOS. [8]

Vitamin D deficiency has been proposed as the possible missing association between insulin resistance and PCOS. This assumption is supported by the finding that the active vitamin D & its re-

ceptor (VDR) complex regulates over 300 genes, including genes that are important for glucose and lipid metabolism as well as blood pressure regulation. [9] Association between Vitamin D status and metabolic and hormonal disorders in women suffering from PCOS is scarce, many studies have concluded that Vitamin D supplementation has increased insulin sensitivity & improvement of PCOS symptoms.

Considering that proper interventions could improve outcomes of PCOS & its related long-term complications, current study aimed to evaluate the effect of vitamin D & calcium supplementation in addition to Metformin to Metformin alone in PCOS women.

Materials and Methods

This is a prospective study done in the Department of Obstetrics and Gynaecology, Katuri Medical College, Guntur for a period of 2 years (Oct 2014-Oct 2016).

This study was designed to compare the efficacy of vitamin D supplementation over Metformin in the management of PCOS with Vitamin D deficiency.

Source of the Study

Women attending to Department of Obstetrics & Gynaecology, Katuri Medical College & Hospital, Chinnakondrupadu, and Guntur with complaints of menstrual irregularities/hirsutism/acne were recruited.

Type of study: Prospective randomised study.

Sample size: 100 subjects with 50 in each Group A & Group B.

Inclusion Criteria:

- Women diagnosed PCOS between ages 15 -35 yrs are included in the study.

Exclusion Criteria:

- Patient not giving consent for the study.
- Patients who have conceived in period of our study.
- Patients with lost follow up.
- Patients taking infertility treatment.
- Patients who underwent any surgical procedures during study.
- Patients with cardiac / renal / hepatic /thyroid disorders & hyperprolactinemia.

Data collection:

Women attending OPD diagnosed as PCOS are recruited into the study & Vitamin D3 levels are estimated. 100 women with diagnosed PCOS and VitaminD deficiency are recruited into the study.

According to endocrine society clinical practice guidelines, Vitamin D levels as vitamin D deficiency as levels of 25OHD D <20ng/mL, vitamin D insufficiency as 25OHD between 21 and 29 ng/mL, vitamin D sufficient as 25OHD level > 30 ng/mL.

Consent was taken from the selected woman after explaining the study procedure.

The detailed history (demography, complaints, menstrual, medical & surgical) was taken & clinical examination (complete general physical examination, systemic) was done.

Investigations like serum Fasting insulin, Trans-abdominal ultrasound for ovarian morphology and volume was done.

Selected women are randomly divided into two groups:

The group A was supplemented with Metformin 1000mg daily along with Vit D3 60k IU weekly for 6 weeks followed by monthly for 4months & Calcium 1000mg daily.

The group B was supplemented only with Metformin 1000mg daily for 6months.

- The cases were followed & response to treatment was assessed (menstrual cycles, hyperandrogenic features, insulin resistance, ovarian morphology & volume) after 6 months.
- The results were compared between the two groups after statistical analysis.
- The data was tabulated and analyzed. Categorical data was analyzed by chi square test and student T test of proportion.
- Multivariate analysis was performed to establish the relationship between Vitamin D3 & Calcium supplementation and outcome along with Metformin in diagnosed Vitamin D3 deficient PCOS women.

Variables measured: Weight, BMI, W-H ratio, Acne, Hirsutism, Serum Vitamin D, Ovarian volume. Serum 25-OH vitamin D was estimated by high performance liquid chromatography (HPLC) method.

The data has been entered into MS-Excel and statistical analysis has been done by using IBM SPSS Version 20.0. To test the association between the groups, chi-square test used. To test the mean difference between two groups, student's t-test (Independent sample t-test / paired sample t-test) was used. To test the correlation between the groups, Pearson's correlation was used. P values are having less than 0.05 are considered as statistical significant.

Results

Table 1: Vitamin D levels in Group A and Group B

Serum 25-OH vitamin D-ng/ml	Group – A		Group – B		P- Value
	(n = 50)	(%)	(n = 50)	(%)	
<10	19	38	10	20	0.4
11-15	17	34	25	50	
16-20	13	26	15	30	
> 21	1	2	0	0	
Mean	12.32 ± 4.37		13.14 ± 4.34		

Table 2: Menstrual abnormalities Before Treatment and after treatment

Before treatment	Group – A		Group – B		P-Value
	(n = 50)	(%)	(n = 50)	(%)	
Amenorrhea	12	24	5	10	0.114
Irregular	17	34	20	40	
Oligo	19	38	25	50	
Regular	2	4	0	0	
After treatment					
Irregular	4	8	13	26	0.031
Oligo	5	10	8	16	
Regular	41	82	28	46	
No change	0	0	1	1	

Table 3: Ovarian Volumes before Treatment and after treatment in Group A and Group B (Volume in Cm³)

Before treatment	Group – A		Group – B		P- Value
	n	%	n	%	
<7	12	24	13	26	0.765
7-10	31	62	30	60	
> 10	7	14	7	14	
Mean	8.62 ± 1.67		8.52 ± 1.65		
After treatment					
	Group – A		Group – B		
	n	%	n	%	
<7	37	74	22	44	<0.0001
7-10	13	26	28	56	
> 10	0	0	0	0	
Mean	6.64 ± 1.064		7.48 ± 1.07		

Table 4: Serum Fasting Insulin (mIU/ml) before Treatment and after treatment in Group A and Group B

before Treatment	Group – A		Group – B		P- Value
	(n = 50)	(%)	(n = 50)	(%)	
<15	7	14	8	16	0.997
16-25	36	72	34	68	
26-35	7	14	8	16	
Mean	19.59 ± 6.23		19.59 ± 6.67		
before Treatment					
	Group – A		Group – B		
	(n = 50)	(%)	(n = 50)	(%)	P- Value
<15	20	40	17	34	0.344
16-25	26	52	28	56	
26-35	4	8	5	10	
Mean	16.01 ± 4.89		17.03 ± 5.77		

Discussion

The management of PCOS depends on the symptoms either ovulatory dysfunction – related menstrual disorders or androgen related symptoms. Correction of BMI and change in life style are the prior steps in the treatment of PCOS. Weight loss improves the endocrine profile & increases the

likelihood of ovulation. Initial weight loss by even 5% can modestly normalize the cycles and ovulation. [10] Low levels of Vitamin D that causing dysregulation of Calcium metabolism may be the primary factors in the initiation & development of PCOS, and the dietary repletion of

this important Vitamin could help to restore normal menstrual cycles in women with PCOS.

In this present study, we evaluated the effect of Metformin and combination of vitamin D, Calcium to Metformin on menstrual regularity, symptoms of hyper androgenism, IR & ovarian volume in 100 PCOS women with vitamin D deficiency after 6 months of supplementation. Mean age distribution in group B (Metformin) of our study was found to be 23.4 yrs. Majority of women with PCOS accounts 72% in group A & 70 % in group B are with severe Vitamin D deficiency with value < 15ng/ dl in both the groups. The mean Vitamin D levels are 12.32 ± 4.37 & 13.14 ± 4.34 in group A & B respectively showing high prevalence of Vitamin D deficiency in PCOS women.

At the admission, majority of women in group A were with oligomenorrhea (38%) & irregular cycles (34%) whereas in group B 50% was with oligomenorrhea & 40% were with irregular cycles, which were similar. At the end of the treatment 82% of women in group A were with regular cycles when compared to only 46% in group B with significant p value 0.031. Thus there was a significant improvement in the regularization of menstrual cycles in group A receiving Metformin & Vitamin D & Calcium when compared to group B receiving only Metformin.

At the admission, majority (62%) in group A & 60% in group B were with ovarian volume between 7 – 10 cm³. The mean volume in group A was 8.62 ± 1.67 & in group B was 8.52 ± 1.65 which is similar. at the end of the treatment the majority of women in group A (74%) have ovarian volume < 7 cm³, whereas majority in group B (56%) had volume between 7-10cm³. The mean volume in group A was 6.64 ± 1.064 & in group B was 7.48 ± 1.07 and the difference in ovarian volume was statistically significant with p value <0.0001 showing addition of Vitamin D & Calcium to Metformin is beneficiary.

At the admission, the Fasting Insulin in majority of women in group A (72%) & in group B (68%) was between 16 – 25 mIU/ml. The mean value in group A was 19.59 ± 6.23 & in group B 19.59 ± 6.67 which is comparable. at the end of the treatment women with Fasting Insulin <15 mIU/ml were 40% in group A & 34% in group B. 52% in group A & 56% in group B were with insulin between 16- 25 mIU/ml. The mean value in group A 16.01 ± 4.89 & group B 17.03 ± 5.77 showed no statistical significance.

The potential mechanisms by which Vitamin D can affect glucose metabolism could be the result of either direct or indirect actions of serum 25OH-D.

- Direct stimulation of insulin release through the expression of VDR as well as the enzyme

1 α -OHase in the pancreatic β cells.

- Through binding of the 1,25 (OH)₂D-VDR complex to the vitamin D response elements of the INSR at the tissue level and thereby enhancing insulin responsiveness for glucose transport.

These are supported by studies showing association between low serum 25OHD levels and increased C-reactive protein levels.

In the present study Vitamin deficient women are 99%, among them 56% are mild deficit with vitamin D levels between 11-20 ng/ml, 34% are moderate deficit with 5-9 ng/ml & 9% are severe deficit with vitamin D levels < 5 ng/ml.

Vitamin D, being a fat-soluble vitamin sequesters in high proportion in adipose tissues lowering the bioavailability in PCOS women specially in obese PCOS women, which leads to high prevalence of Vitamin D deficiency in PCOS women [11].

Calcium homeostasis is regulated by Vitamin D, by altering Calcium signaling pathway increases in Calcium influx. This helps in activation & maturation of oocytes in turn results in progression of follicular development & ovulation leading to normalization of menstrual cycles. [11,12]

The present study showed a significant improvement by 30% in reduction of ovarian volume after treatment, which indirectly signifies the more chances for maturation of follicles.

In the present study, while comparing the efficacy between two groups, there is 18.4% in group A & 13 % in group B reduction in the mean serum Fasting insulin showing there is no significant differences in outcome i.e reducing the serum fasting insulin on addition of Vitamin D & Calcium to Metformin. [12]

Conclusion

PCOS is the most common endocrine problem affecting the women from menarche to perimenopausal age. Vitamin D deficiency is very common in PCOS. A variety of treatment strategies have been developed based on the patient symptomology & need. Insulin sensitizers specially Metformin has proven efficacy in treatment of PCOS since long time. Vitamin D & Calcium supplementation in addition to Metformin has shown potential therapeutic benefits in ameliorating the hormonal milieu and a variety of PCOS related symptoms mainly menstrual regularity, ovulation & some features of hyperandrogenism. Thus, Vitamin D & Calcium is a safe drug with less side effects & thus recommended for these group of women especially those with Vitamin D deficiency.

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