Bangladesh J Obstet Gynaecol, 2024; Vol. 39(2): 104-110

Vitamin-D in Women with Primary Infertility in a Tertiary Care Centre in the Southeast Region of Bangladesh

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Abstract:

Introduction: *Infertility affects 9–18% of the population, with vitamin D deficiency being a potential contributing factor. This study evaluates the association between vitamin D levels and primary Infertility in women attending a tertiary care centre in Southeast Bangladesh.*

Methods: *A cross-sectional study was conducted at Chittagong Medical College Hospital's(CMCH) Infertility Unit over one year(February 2020 to January 2021). Ninety-two women with primary Infertility were recruited based on specific inclusion criteria. Data collection included demographic details, clinical history, hormonal profiles, and vitamin D levels measured via Chemiluminescent Microparticle Immunoassay. Vitamin D status was classified according to the 2011 Endocrine Society guidelines. Statistical analysis was performed using SPSS version 23.*

Results: *All the participant's Vitamin D level were found less than normal.The majority (75%) of women had insufficient (<30 ng/ml) vitamin D levels, with 14.1% classified as severely deficient (<10 ng/ml). Vitamin D deficiency was significantly associated with urban residence and limited sun exposure (p=0.021). Clinical characteristics revealed 45.7% of participants had PCOS, and a significant portion displayed obesity (BMI ≥25 kg/m²). Hormonal analysis highlighted elevated LH levels in 44.8% and low AMH in 13.5%.*

Discussion: *Vitamin D deficiency was highly prevalent among infertile women, supporting its potential link to reproductive dysfunctions such as ovulatory disorders. PCOS and metabolic abnormalities further compounded infertility risks. These findings align with global studies, suggesting vitamin D supplementation as a promising adjunct in infertility management.*

Conclusion: *Vitamin D deficiency is a significant concern in women with primary Infertility in Bangladesh. The study emphasizes the need for targeted interventions, including supplementation and lifestyle modifications, to improve reproductive outcomes.*

Keywords: *Vitamin D, primary Infertility, PCOS, reproductive health.*

Introduction

Infertility has become a significant concern in recent times, affecting 9%-18% of the general population, and is a common condition causing distress among couples[1]. It is a complex medical disorder that impacts the economic and psychological well-being of those affected[2]. The causes of female Infertility are varied, including genetic and anatomic abnormalities, endocrine issues, and autoimmune disorders[3-6]. However, unexplained Infertility accounts for nearly 50% of cases, with substantial Evidence suggesting that a portion of these cases may be directly or indirectly linked to autoimmunity[7-8].

Vitamin D, a steroid hormone, is well-known for its role in maintaining calcium homeostasis and promoting bone mineralization[9]. Recent Evidence also highlights the non-skeletal benefits of vitamin D, such as its correlation with chronic conditions including

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endocrine diseases[3-6], cancer development and progression[7-8] and autoimmune disorders[10]. As a positive immune modulator, vitamin D influences both the innate and adaptive immune responses[11], with hypovitaminosis D being prevalent among women and associated with disease activity and comorbidities[11].

Vitamin D plays a significant role in reproductive processes in both men and women due to the expression of vitamin D receptors (VDR) and 1-alpha-hydroxylase in reproductive tissues, including the ovaries, testes, uterus, placenta, pituitary gland, and hypothalamus[11-13]. Approximately 80-90% of vitamin D is synthesized in the skin via sunlight exposure. At the same time, the remainder is obtained from food sources[14]. Seasonal variations in serum vitamin D levels have been observed, with higher levels in summer compared to winter, potentially influencing natural conception and birth rates[15-16].

Vitamin D exists in two isoforms: vitamin D3 (cholecalciferol) and vitamin D2 (ergocalciferol)[17]. w Its metabolite, 25-hydroxyvitamin D 25(OH)D, is produced in the liver, with a serum half-life of 2-3 weeks. In circulation, 85-90% of 25 (OH) D binds to vitamin D-binding protein (VDBP), 10-15% to albumin, and less than 1% circulates freely to exert cellular effects[18]. Despite the lack of consensus on optimal vitamin D levels, serum levels of 30 ng/ml are generally considered sufficient[19].

Vitamin D deficiency is frequently diagnosed in women of reproductive age, with rising prevalence attributed to obesity, lifestyle factors, and reduced sun exposure[20-23]. Studies suggest that the ovary may be a target organ for 1,25(OH), Dƒ , possibly influencing ovarian activity[24]. Evidence from animal and human studies underscores the importance of vitamin D in human reproduction, with reports of high prevalence of vitamin D deficiency among women undergoing in vitro fertilization[25-26].

Clinical studies have demonstrated a positive correlation between adequate vitamin D levels and successful fertility treatments in women with Infertility[25]. It has been suggested that vitamin D may enhance ovarian steroidogenesis and improve endometrial receptivity[25-28].

Vitamin D deficiency has been extensively studied as a potential cause of Infertility. However, there is no consensus regarding its precise impact on reproductive health. While optimal vitamin D levels are considered essential for managing conditions like PCOS, endometriosis, male Infertility, and IVF outcomes, no definitive correlation has been established between vitamin D levels and ovulation stimulation or embryo development. This study aims to assess serum vitamin D levels in women presenting with primary Infertility at a tertiary care centre in the southeast region of Bangladesh, exploring potential therapeutic approaches like vitamin D supplementation.

Methods:

This study was a cross-sectional analysis conducted in the Infertility Unit of the Department of Obstetrics and Gynecology at Chittagong Medical College Hospital (CMCH) over one year (From February 2020 to January 2021) following ethical clearance from the CMCH Ethical Review Committee (Memo No.: CMC/PG/2022/233; 19/01/2020). Eligible participants were women of reproductive age diagnosed with primary Infertility. Women were excluded if they had known liver, kidney, or bone diseases, severe infections, congenital uterine anomalies, tubal or cervical factor infertility, male factor infertility, or if they were on vitamin D supplementation.

A total of 92 women of reproductive age, presenting with primary Infertility of varying etiologies, were recruited for the study. Participants were selected using non-probability sampling. Data were collected using a structured case record form that included a detailed questionnaire. Blood samples were taken from each participant on their first consultation day, regardless of the menstrual cycle day. Additional hormonal assessments, including FSH and LH, were performed on the 2nd or 3rd day of the menstrual cycle, with other tests such as TSH, serum prolactin, and AMH conducted irrespective of the cycle day. Antral follicle counts were assessed using transvaginal ultrasonography (TVS) on cycle days 1–5.

Blood samples were collected in plain serum tubes, centrifuged within one hour, and analyzed using Chemiluminescent Microparticle Immunoassay (CMIA). Vitamin D 25(OH)D levels were graded as per the 2011 Endocrine Society guidelines:

- Insufficient: 20-29 ng/ml
- Deficiency: <20 ng/ml
- Severe Deficiency: <10 ng/ml

Participant's body mass index (BMI) was calculated in kg/m². Physical activity levels, sun exposure, and clinical histories were recorded.

Categorical variables were expressed as frequencies and percentages, while continuous variables were presented as means ± standard deviations (SD). Statistical analysis was performed using SPSS version 23.0, with a chi-square test employed for comparison. P-values <0.05 were considered statistically significant.

Result::

In this study, all the participant's Vitamin D levels were found to be less than normal(95.7%).Table I summarizes the demographic and Infertility characteristics of 92 women categorized by their serum 25-hydroxyvitamin D (25OH-D) levels into two groups: <20 ng/ml (n=69) and ≥20 ng/ml (n=23). The majority of participants were aged 18-25 years, and a similar distribution was observed across other age groups. Most participants were Muslim (88.0%), with no significant difference in religious distribution. A notable difference was found in residence, where a higher percentage of women with ≥20 ng/ml 25OH-D lived in rural areas (65.2%) compared to those with <20 ng/ml (37.7%). Educational levels and socio-economic status were similar across both groups, with the majority having HSC and above education and being from the middle class. Seasonal distribution showed most women were evaluated during the winter months, with no significant seasonal variation in vitamin D levels.

The table-II presents data on various factors related to the infertile group of women (N=92). Regarding physical activity, 58.7% reported being inactive, while 41.3% engaged in some form of activity. The majority experienced Infertility for 1-5 years (79.3%), with fewer cases reporting longer durations. Most women had a body mass index (BMI) in the 25-30 kg/m² range (47.8%), with 34.8% in the 19-25 kg/m² range, indicating a significant prevalence of overweight and obesity. Sun exposure was reported by 33.7%, while 66.3% did not get daily sun exposure. The group was evenly split regarding menstrual cycle regularity, with 50% having regular and 50% having irregular cycles. The duration of marriage for most participants was less than 5 years (65.2%). A large majority (88.0%) had no history of contraception use, with only 12.0% reporting previous use.

Table III presents the distribution of vitamin D levels among 92 women with primary Infertility. The mean

Table-I

Demographic profile and infertility characteristics of cases, infertile group (N=92)

Variables	25-OH-D		Frequency (%)	P value
	<20ng/ml [Deficiency] (n=69)	≥20ng/ml [Insufficiency] (n=23)		
Age groups, years				
18-25	36 (52.2)	14 (60.9)	50(54.3)	0.317
26-30	26 (37.7)	5 (21.7)	31(33.7)	
>30	7 (10.1)	4 (17.4)	11(12)	
Religion				
Muslim	61 (88.4)	20 (87.0)	81(88.0)	0.853
Others	8 (11.6)	3 (13.0)	11(12.0)	
Residence				
Rural	26 (37.7)	15 (65.2)	41(44.6)	0.021
Urban and Semi-urban	43 (62.3)	8 (34.8)	51(55.4)	
Education				
Primary or bellow	14 (20.3)	3 (13.0)	17(18.4)	0.447
SSC	13 (18.8)	7 (30.4)	20(21.7)	
HSC and above	42 (60.9)	13 (56.5)	55(59.8)	
Socio-economic condition				
Upper class	2 (2.9)	1 (4.3)	3(3.3)	0.831
Middle class	63 (91.3)	20 (87.0)	83(90.2)	
Lower class	4 (5.8)	2 (8.7)	6(6.5)	
Season				
Winter(Dec-Feb)	56 (81.2)	19 (82.6)	75(81.5)	0.592
Summer (March-May)	3 (4.3)	2 (8.7)	5(5.4)	
Monsoon (June-Sept)	10 (14.5)	2 (8.7)	12(13)	

(*) The result is expressed as frequency and percentages & Chi-square test.

Table-II

Clinical characteristics of 92 women with primary Infertility

	Frequency	Percentage
Physical activity		
Absent	54	58.7
Present	38	41.3
Duration of Infertility (years)		
1-5	73	79.3
6-10	13	14.1
11-15	5	5.4
>15	1	1.1
Body mass index, kg/m^2		
<18	1	1.1
19-25	32	34.8
25-30	44	47.8
>30	15	16.3
Exposure to sun/daily		
Yes	31	33.7
No	61	66.3
Menstrual cycle		
Regular	46	50.0
Irregular	46	50.0
Duration of marriage (Years)		
<5	60	65.2
6-10	22	23.9
11-15	10	10.9
H/O contraception		
Yes	11	12.0
No	81	88.0

(*)The result is expressed as frequency and percentages.

Table-III

Vitamin D levels in women with primary Infertility (N = 92)

Vitamin D	Frequency	Percentage
Mean ±SD, ng/ml	16.04±6.80	
Range, ng/ml	4.20-41.14	
Status		
Severe deficient <10 ng/ml)	13	14.1
Deficient (10.0-19.9 ng/ml)	56	60.9
Insufficient (20.0-29.9 ng/ml)	19	20.7
Sufficient (≥30.0 ng/ml)	4	4.3

vitamin D level was 16.04 ng/ml with a standard deviation of 6.80 ng/ml, and levels ranged from 4.20 to 41.14 ng/ml. The majority of women (60.9%) were categorized as deficient (10.0-19.9 ng/ml), while 14.1% were severely deficient (<10 ng/ml). Additionally, 20.7% of the participants had insufficient vitamin D levels (20.0-29.9 ng/ml), and only a small percentage (4.3%) had sufficient levels (≥30.0 ng/ml). This distribution highlights a high prevalence of vitamin D deficiency among the study population.

The Figure-1 outlines the presenting complaints among women with primary Infertility, highlighting various symptoms experienced. The most commonly reported symptom was weakness, affecting 33.7% of the women. Muscle aches were reported by 22.8%, while bone pain affected 25.0% of the participants. Additionally, 23.9% of the women experienced depression and headaches, indicating a significant overlap in psychological and physical symptoms. Profuse sweating was reported by 11.9% of the women, the least common complaint. These findings emphasize the diverse range of symptoms associated with primary Infertility and vitamin D deficiency, including both physical discomforts and mental health challenges.

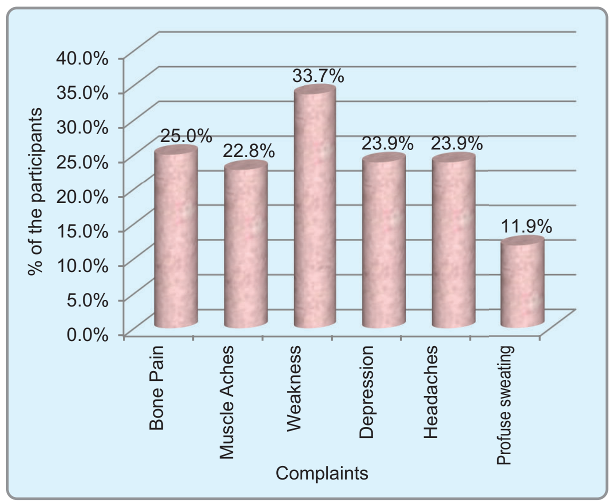

Figure-I: *Presenting complaints of the women with primary Infertility*

The medical conditions associated with primary Infertility in women highlight that polycystic ovary syndrome (PCOS) is the most prevalent, affecting 45.7% of participants. Hirsutism follows as the second most common condition at 25.0%. Other conditions reported include endometriosis at 8.7% and diabetes mellitus at 3.3%. Additionally, less common conditions such as fibroids and hypertension each affect 2.2% of the women, while premature ovarian failure, adenomyosis, hyperprolactinemia, and pelvic inflammatory diseases each affect 1.1% to 1.2%. Overall, the data illustrate a diverse range of gynecological and systemic conditions linked to primary Infertility.

Table-IV presents ovarian reserve parameters and other biochemical markers in women with primary Infertility. Among the 37 women assessed for Anti-Müllerian Hormone (AMH), 59.5% had levels above 3 ng/ml, indicating a good ovarian reserve, while 13.5% had low levels (<1 ng/ml). Follicle-stimulating hormone (FSH) levels were within the normal range (1.3-9.9 miu/ml) in 94.4% of the 54 women tested. Luteinizing Hormone (LH) levels were found to be greater than 6 miu/ml in 44.8% of the 58 women, suggesting potential ovulatory dysfunction. Antral Follicle Count (AFC), assessed in 40 women, revealed that 65.0% had a count of 6-15, indicating a normal ovarian reserve, while 32.5% had a count above 15. Serum prolactin levels were normal (<30 miu/ml) in 75.0% of the 72 women tested, with elevated levels (>50 miu/ml) in 5.6%.

Table-IV

Ovarian reserve parameters and other biochemical parameters of the women with primary Infertility

Parameters	Frequency	Percentage
AMH, ng/ml (n=37)		
<1	5	13.5
1.1-3	10	27.0
>3	22	59.5
FSH, miu/ml (n=54)		
<1.2	1	1.9
1.3-9.9	51	94.4
>10	2	3.7
LH, miu/ml (n=58)		
<1.2	2	3.4
1.3-6	30	51.7
>6	26	44.8
AFC (n=40)		
<5	1	2.5
6-15	26	65.0
>15	13	32.5
Serum prolactin, miu/ml(n=72)		
<30	54	75.0
30-50	14	19.4
>50	4	5.6
Serum TSH miu/ml (n=79)		
<1	3	3.8
1.1-3.5	57	72.2
>3.5	19	24.1

Thyroid-stimulating hormone (TSH) levels were within the normal range (1.1-3.5 miu/ml) in 72.2% of the 79 women, while 24.1% had elevated TSH levels, indicating potential thyroid dysfunction.

Discussion:

The study thoroughly examines the clinical, biochemical, and demographic profiles of women with primary Infertility. It highlights noteworthy observations on ovarian reserve parameters, related medical disorders, and vitamin D deficiency. The prevalence and features of diseases related to Infertility exhibit both similarities and differences when these results are compared to those of earlier investigations.

In this study, 75% of individuals had vitamin D levels that were either insufficient (20–29.9 ng/ml) or deficient (<20 ng/ml), indicating a high incidence of vitamin D deficiency among women with primary Infertility. This is in line with research which found that vitamin D deficiency is prevalent among infertile women and may have a negative impact on the success of their reproductive efforts.[12] In addition, another study reaffirmed the significance of vitamin D for reproductive health, pointing out that supplementation may enhance ovarian function and monthly regularity of menstruation and ovulation.[29] Mechanistic evidence linked vitamin D insufficiency to decreased ovulatory function in a review. Insufficient vitamin D was linked to reproductive hormone imbalance. [30] Vitamin D insufficiency was detected in 64.28% of infertile Indian women.[31]

The study found that 45.7% of women with Infertility had PCOS, which is consistent with global estimates of 40–50% prevalence among these women, according to a comprehensive analysis.[32] In keeping with findings by Yildiz et al. (2018) that highlight the comorbid nature of Infertility with metabolic and endocrine illnesses, the study also found other conditions like endometriosis, hirsutism, and metabolic disorders.[33]

Anti-Müllerian Hormone (AMH) levels were assessed in the study population, and the results showed that most had normal levels; only 13.5% had low levels, which may indicate reduced ovarian reserve. The results are in line with another study which talked about the function of AMH as a trustworthy indicator of ovarian reserve and how it relates to reproductive age.[34] The results of the study on LH and FSH levels also shed light on the study population’s endocrine profile, which is crucial for comprehending the underlying pathophysiology of Infertility. A PCOS case-control research found that women with 25(OH)D3 deficiency had significantly higher levels of testosterone, dehydroepiandrosterone-sulfate, and LH ($p<0.05$). In addition,25(OH)D3 concentrations were adversely linked with body mass index ($r=-0.459$), serum testosterone ($r=-0.374$), and dehydro-epiandrosterone-sulfate levels ($r=-0.418$; both; $p<0.05$).[35]In an Indian study, in infertile females with vitamin D deficiency, the mean AMH was found to be 1.94 ± 1.30.[31]

The study's conclusions align with worldwide research patterns, suggesting that infertile groups share common etiological variables such as vitamin D insufficiency(25(OH)D3), PCOS, and endocrine problems. Research conducted in 2019 also brought attention to the multidimensional nature of Infertility, which includes aspects related to lifestyle, metabolism, and hormones.[36]

Conclusion:

This study underscores the significant prevalence of vitamin D deficiency among women with primary Infertility in Bangladesh, with the majority of participants presenting insufficient or deficient levels. The findings reinforce existing evidence linking vitamin D deficiency to reproductive issues, including ovulatory dysfunction and hormonal imbalances. Notably, the high prevalence of PCOS further highlights the complex interplay between vitamin D status, metabolic health, and reproductive function. Given the potential therapeutic benefits, vitamin D supplementation and lifestyle interventions may serve as promising strategies to improve fertility outcomes. Further, longitudinal studies are warranted to establish a causal relationship and evaluate the efficacy of targeted interventions in managing Infertility

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