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Research Article

Association of Vitamin D Status with Semen Parameters and Male Reproductive Hormones in Egyptian Infertile Men

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Abstract

Background: Although vitamin D receptor (VDR) is expressed in human sperm, little is known about the role of vitamin D (Vit. D) in male reproduction. Our objective was to assess Vit. D levels both in serum and seminal fluid and to establish the relation between serum and seminal Vit. D levels, semen parameters, male sex hormones and serum calcium level in Egyptian infertile men.

Patients and Methods: We conducted a prospective case control study including 30 healthy fertile males as a control group and 60 male patients with infertility of unknown etiology. Semen samples were collected and semen parameters were evaluated. Also, seminal Vit. D level was measured. Blood samples were taken as serum levels of Vit. D, calcium, testosterone, Luteinizing Hormone (LH) and Follicle-Stimulating Hormone (FSH) were estimated.

Results: There was significant decrease of both serum and seminal Vit. D level in groups of male infertility compared to control group. A significant positive correlation was found between serum and seminal Vit. D levels in different study groups. Also, significant positive correlation between serum Vit. D level and non-progressive sperm motility.

Conclusion: Our results support the role of Vit. D in semen parameters and male fertility status.

Keywords: Vitamin D; Male infertility; Semen parameters; Reproductive hormones

Background

Infertility is failure to achieve a clinical pregnancy after a year or more of unprotected and regular sexual intercourse [1], or due to an impairment of a person's capacity to reproduce either as an individual or with his/her partner. There are 2 types of male infertility. Primary male infertility which means a male who has never initiated a clinical pregnancy and meets the criteria of being classified as infertile. While a male who is unable to initiate a clinical pregnancy, but had previously initiated a clinical pregnancy is defined as secondary male infertility [2]. About 15% of couples are suffering from infertility, among them; approximately 50% of the cases infertility is partially or wholly attributable to a male factor [3,4].

Male infertility can be due to identifiable hormonal or anatomical causes that may be reversible or irreversible [5] Hormonal etiologies of male infertility are often referred as pre-testicular causes. Fertility impairment in these cases is due to either hormonal deficiency or excess [6]. Normal functioning of the hypothalamus, pituitary glands and testes is required for male fertility and complete male germ cell maturation is dependent on the balanced hormonal secretion of these glands [7].

Hypogonadotrophic hypogonadism which is the failure of pituitary gland to secrete adequate amounts of FSH and LH can lead to decreased sperm count and male infertility [8]. In addition; in oligospermic males hyperprolactinemia can cause infertility. It inhibits the pulsatile secretion of the gonadotrophin releasing hormone with subsequent decreased pulsatile release of FSH, LH and testosterone, which in turn causes spermatogenic arrest and impaired sperm motility. It leads to secondary hypogonadism and male infertility [9].

The exact etiology of male infertility remains unknown in 30% to 50% of patients, who are classified as having idiopathic male infertility [10], which characterized by the presence of abnormal semen parameters without a discernible cause and absence of female infertility [11]. In contrast, unexplained male infertility is the inability of the male to establish pregnancy, despite having normal semen parameters without any known cause of infertility and the absence of female factor of infertility [12].

Although vitamin D (Vit. D) is most strictly connected with regulation of calcium and bone homeostasis [13], it has been suggested to have many other actions, including effects on the immune system, diabetes, and cancer prevention [14,15]. One of the recently identified target zones of Vit. D is male reproductive function. The basis of the interplay between Vit. D and reproduction lays on the expression of both Vit. D in Vit. D receptors (VDR) and all the Vit. D3 metabolic enzymes (CYP2R1, CYP27B1, and CYP24A1) in different tissues

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of the reproductive system in both sex [16]. VDR was shown to be expressed in neck and post-acrosomal regions of the sperm, and found to be higher in normal men than in infertile men [17]. The metabolizing enzymes are expressed in testis, epididymis, seminal vesicle, prostate, and spermatozoa suggesting a local regulation of active Vit. D that may be important for spermatogenesis and sperm function [18].

The exact role of VDR in the sperm nucleus is not known. It may act as a protective genomic factor, as it is essential for the proper control of sperm DNA integrity and maintenance of genome stability [19]. Vit. D could decrease early apoptosis and necrosis, and increase sperm motility in asthenozoospermia [20]. Hypovitaminosis D has a negative impact on semen and hormone function, either in animals or in humans [21].

It has been suggested that Vit. D increases intracellular calcium concentration and sperm motility, and induces acrosome reaction in mature sperm from healthy males [18,22]. Vit. D might also enhance sperm motility by promoting the synthesis of ATP, both through the cAMP/PKA pathway and the increase in intracellular calcium ions [23,24].

Previous researchers have found positive relationship between the serum level of Vit. D and semen parameters [18-22]. However, the relation between serum and seminal levels of Vit. D, semen parameters and male sex hormones, is still unclear.

Our objective was to assess both seminal and serum Vit. D levels in Egyptian infertile males and to verify if serum Vit. D level is a reflection of its seminal level. Also, to establish the relation between serum and seminal Vit. D levels, semen parameters, male reproductive hormones and serum calcium level in Egyptian infertile men.

Patients and Methods

Study design and population

A prospective case control study was performed at Dermatology, Venereology and Andrology Department and Medical Biochemistry Department, Faculty of Medicine, Assiut University, Assiut, Arab Republic of Egypt. The study received an approval from the Medical Ethics Committee of Faculty of Medicine, Assiut University (Approval number IRB17100087). From all participants an informed consent was obtained before the study.

All infertile male patients attending the Andrology Outpatient Clinic of Assiut University Hospital (AUH) were screened. After excluding 186 cases of infertile men not fulfilled the inclusion criteria, a total of 60 male patients with infertility of unknown etiology were incorporated in the study. In addition, 30 healthy fertile males (as controls) were randomly recruited.

Inclusion criteria included infertile male patients without any detectable cause of infertility with normal clinical examination ± normal semen parameters.

Exclusion criteria

The study excluded patients with any apparent physical finding and any known pathology of the reproductive tract (e.g. prostatitis, epididymitis, any genital tract infections, varicocele, diminished testicular volume or abnormal hormonal profile, etc.), or those received Vit. D therapy or other hormonal therapy. Moreover, patients with chronic systemic disease, cancer, malabsorption, poor general status or combined male and female factor of infertility were also excluded.

The participants were subdivided into 3 groups

- Control group: included 30 fertile males.
- Unexplained male infertility group: included 30 infertile male patients with normal clinical examination and normal semen parameters.
- Idiopathic male infertility group: included 30 infertile male patients presented with normal clinical examination and abnormal semen parameters.

A detailed history was taken from all patients including: personal, sexual, medical history and family history in addition to the fertility history of his wife. General physical examination and genital examination of testis, epididymis, vas deferens, spermatic cord, penis, scrotum and inguinal lymph nodes were performed.

Semen analysis

From all patients semen samples were collected after 2-5 days of sexual abstinence. It was obtained by masturbation into a sterile plastic container. Patients were instructed to report any semen loss during semen collection. After complete liquefaction in 37°C incubator, semen samples were evaluated for physical criteria and then centrifuged where the pellets are used to assess the semen parameters based on World Health Organization (WHO) laboratory manual [3]. Normal sperm parameters are considered if total sperm count >39 million sperm per ejaculate, sperm concentration >15 million sperm per ml, progressive motility >32%, total motility >40% and normal morphology ≥4%. Seminal fluid was evaluated for Vit. D level as described in the kit manual supplied by Epitope Diagnostics, Inc (EDITM Total 25-OH Vit. D EIA Kit, Cat No.: KT715, United States).

Biochemical analysis

Venous blood samples from all patients were collected *via* venipuncture of superficial vessels in the antecubital fossa or hands by a well-trained clinician. Five ml of peripheral venous blood was withdrawn from each patient; it was dispensed into plain tubes then centrifuged for 15-minutes at the speed of 2000-3000 r.p.m. and serum collected. Serum is divided into 5 eppendorf tubes then stored in -20°C to avoid repeated freeze-thaw cycles till batch assay of our markers. Serum level of total Vit. D was measured using the same kit as seminal levels (EDITM Total 25-OH Vit. D EIA Kit, Cat No.: KT715, United States), serum calcium was measured by colorimetric method using abcam assay kit (ab102505, United Kingdome). FSH and LH levels were measured by enzyme-linked immunosorbent assay (ELISA) kits supplied by Perfecta Ease Biotec (Beijing) co., USA, Cat No.:10001 and 10004 respectively. Testosterone Hormone levels were detected by ELISA kit supplied by Abia (AB diagnostic system, German, Cat No.: DK.004.01.3).

Statistical analysis

The data were analyzed using the Statistical Package of Social Science (SPSS version 19; SPSS Inc., Chicago, IL) software program. Data statistically stated in terms of number, percentage, mean,

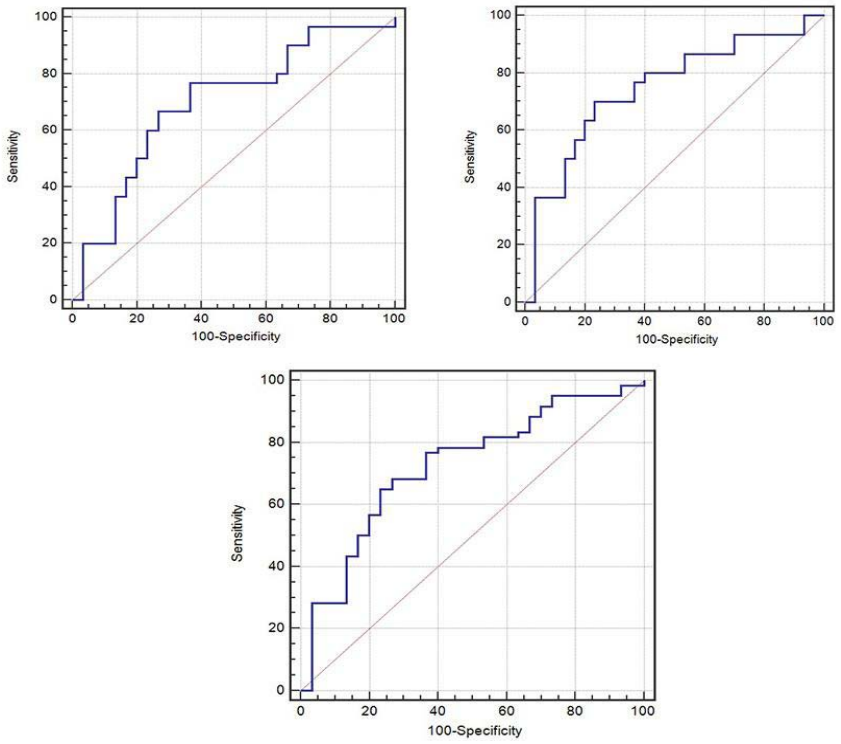

Figure 1: ROC curve to distinguish the cut-off value of serum Vit. D of unexplained infertility, idiopathic infertility and infertility as whole.

median, range and Standard Deviation (SD). Chi-square test was used to compare between qualitative variables. Independent samples t-test was used to compare quantitative variables between two groups in case of parametric data and Mann-Whitney test was used for non-parametric data. Spearman correlation was used to measure the correlation between quantitative variables. A P-value ≤0.05 was considered statistically significant.

Determination of the threshold value for optimal sensitivity and specificity of our markers was done by the Receiver Operating Characteristics (ROC) curve, which was plotted by calculating sensitivity and specificity at multiple cut-off points. Area Under the Curve (AUC) of the ROC plots was calculated using discriminate analysis, where AUC=1.0 means perfect separation of test values into two groups and AUC = 0.5 means no distributional differences. AUC >0.8 indicates excellent discriminating power of the test and AUC >0.7 indicates a discriminating strength of statistical significance.

Results

The Demographic data of the study

The demographic data of the studied groups were summarized in (Table 1). A total of 60 infertile male patients of unknown etiology were included in the study and were divided into 2 groups as mentioned before. There was no statistical significant difference in the mean age between the studied case groups and the control healthy group (29.93±4.39, 29.20±5.27 versus 28.70±3.71 respectively). Both types of infertility were more common in smokers, workers and rural residents.

A statistically significant difference was found between the two diseased groups regarding the infertility duration as the idiopathic male infertility group suffered from a longer period of infertility (1-8 years) in comparison to the unexplained group (1-4 years) (p=0.000).

Characteristics of participants' semen analysis

Table 2 showed significant decrease of semen volume in idiopathic male infertility group compared to both control and unexplained male infertility group (p=0.000). The two male infertility groups showed a significant decrease in sperm count and concentration compared to control group, where the idiopathic male infertility group have lower sperm count and concentration compared to unexplained male infertility group.

As regards, the sperm motility (progressive and non-progressive), there was a significant decrease in both infertility groups compared to control group and also in idiopathic male infertility group compared to unexplained group. In addition, the idiopathic male infertility group expressed a significant decrease in sperm morphology compared to both control and unexplained male infertility group (p=0.000).

Characteristics of participants' hormonal profile

The reproductive hormonal profile revealed no significant among the three groups except for testosterone, where its levels showed a significant decrease in unexplained male infertility group compared to control group (p=0.005). Serum calcium levels were almost equal in different study groups. There was significant decrease in serum and seminal Vit. D level in both unexplained and idiopathic male infertility group compared to control group (p=0.008, p=0.001, p=0.000, p=0.0000 respectively (Table 3).

Moreover, seminal Vit. D level was significantly lower in idiopathic male infertility in comparison to unexplained group, this was shown in (Table 3).

Table 1: Demographic data of the studied participants.

	Control		Unexplained infertility		Idiopathic infertility		P-value[1]	P-value[2]	P-value[3]
	n	%	n	%	n	%			
Age (years)							0.245	0.679	0.571
Mean ± SD	28.70 ± 3.71		29.93 ± 4.39		29.20 ± 5.27				
Median (Range)	28 (23-39)		30 (19-37)		30 (24-39)				
Smoking							0.301	0.07	0.091
Smoker	14	46.70%	18	60.00%	16	53.30%			
Non-smoker	16	53.30%	12	40.00%	14	46.70%			
Residence							0.405	0.037*	0.197
Rural	19	63.30%	22	73.30%	26	86.70%			
Urban	11	36.70%	8	26.70%	4	13.30%			
Occupation							0.076	0.000*	0.014*
Employee	13	43.30%	5	16.70%	0	0.00%			
Farmer	1	3.30%	1	3.30%	6	20.00%			
Worker	16	53.30%	24	80.00%	24	80.00%			

*P-value ≤0.05 is significant.
P[1]: Unexplained infertility versus control,
P[2]: Idiopathic infertility versus control,
P[3]: Unexplained infertility versus Idiopathic infertility.

Table 2: Semen parameters in different study groups.

	Control	Unexplained infertility	Idiopathic infertility	P-value[1]	P-value[2]	P-value[3]
Volume (ml)				0.494	0.000*	0.003*
Mean ± SD	3.10 ± 0.81	2.98 ± 0.99	2.27 ± 0.55			
Median (Range)	3.0 (2.0-4.0)	3.0 (1.5-5.0)	2.0 (1.5-3.0)			
Sperm concentration (10^6/ ml)				0.002*	0.000*	0.000*
Mean ± SD	66.13±21.10	48.13±20.96	16.93±15.80			
Median (Range)	70 (28-100)	45 (20-90)	11 (1-45)			
Sperm count (10^6/ ejaculate)				0.001*	0.000*	0.000*
Mean ± SD	207.33±94.15	145.10±93.06	38.70±34.00			
Median (Range)	192 (70-400)	120 (40-400)	31.5 (1.5-95)			
Rapid linear progressive motility				0.028*	0.000*	0.000*
Mean ± SD	22.33 ± 9.07	16.83 ± 8.35	2.07 ± 3.34			
Median (Range)	20.0 (10.0-40.0)	17.5 (5.0-30.0)	0.0 (0.0-10.0)			
Slow & non-linear progressive motility				0.017*	0.000*	0.000*
Mean ± SD	35.83 ± 9.57	30.50 ± 8.34	14.23 ± 7.31			
Median (Range)	37.5 (15.0-50.0)	27.5 (20.0-50.0)	15.0 (0.0-25.0)			
Progressive motility				0.000*	0.000*	0.000*
Mean ± SD	58.17 ± 9.42	47.33 ± 10.89	16.30 ± 8.12			
Median (Range)	55.0 (40.0-70.0)	45.0 (35.0-70.0)	15.0 (0.0-30.0)			
Non-progressive Motility				0.084	0.000*	0.000*
Mean ± SD	22.83 ± 5.20	25.83 ± 8.21	10.40 ± 5.11			
Median (Range)	22.5 (15.0-30.0)	25.0 (10.0-40.0)	10.0 (1.0-20.0)			
Total motility				0.010*	0.000*	0.000*
Mean ± SD	81.00 ± 8.75	73.17 ± 12.00	26.70 ± 10.18			
Median (Range)	80.0 (65.0-95.0)	75.0 (50.0-90.0)	30.0 (3.0-35.0)			

Immotile sperms						
Mean ± SD	18.83 ± 8.78	26.83 ± 12.00	73.30 ± 10.18	0.008*	0.000*	0.000*
Median (Range)	17.5 (5.0-35.0)	25.0 (10.0-50.0)	70.0 (65.0-97.0)			
Sperm morphology						
Mean ± SD	5.83 ± 1.39	5.37 ± 1.22	2.20 ± 1.13	0.191	0.000*	0.000*
Median (Range)	6 (4-8)	5 (4-8)	2 (1-5)			

*P-value ≤0.05 is significant.
P[1]: Unexplained infertility versus control,
P[2]: Idiopathic infertility versus control,
P[3]: Unexplained infertility versus Idiopathic infertility.

Table 3: Distribution of serum levels of male reproductive hormonal, serum calcium level and serum and seminal vitamin D levels in different study groups.

	Control	Unexplained infertility	Idiopathic Infertility	P-value[1]	P-value[2]	P-value[3]
FSH (mIU/ml)						
Mean ± SD	5.76 ± 1.18	6.26 ± 2.74	6.07 ± 2.41	0.888	0.615	0.745
Median (Range)	5.5 (4.1-8.2)	5.3 (4.0-15.3)	5.3 (3.9-13.9)			
LH (mIU/ml)						
Mean ± SD	6.36 ± 1.25	6.40 ± 0.78	6.26 ± 0.95	0.455	0.918	0.231
Median (Range)	6.2 (4.9-9.8)	6.2 (5.0-8.9)	6.0 (4.5-8.6)			
Testosterone (ng/dl)						
Mean ± SD	491.67±83.02	431.29 ± 62.52	453.29±56.69	0.005*	0.085	0.217
Median (Range)	474 (322-668)	422 (285-523)	460 (301-589)			
Calcium (mg/dl)						
Mean ± SD	7.39 ± 0.72	7.30 ± 0.76	7.37 ± 0.75	0.433	0.446	0.663
Range	5.1-8.5	6.3-9.0	6.2-9.1			
Serum Vitamin D (ng/ml)						
Mean ± SD	24.08 ± 4.58	21.46 ± 3.90	20.24 ± 4.57	0.008*	0.001*	0.188
Range	8.9-30.4	16.2-31.6	14.6-30.2			
Seminal Vitamin D (ng/ml)						
Mean ± SD	11.23 ± 1.30	8.39 ± 1.51	7.31 ± 1.33	0.000*	0.000*	0.003*
Range	8.4-13.9	5.9-10.9	5.6-10.1			

*P-value ≤0.05 is significant.
P[1]: Unexplained infertility versus control,
P[2]: Idiopathic infertility versus control,
P[3]: Unexplained infertility versus Idiopathic infertility.

Correlations between serum and seminal Vit. D levels, semen parameters, male reproductive hormones and serum calcium level in different groups

A significant positive correlation was found between serum and seminal Vit. D levels in unexplained and idiopathic male infertility groups (r=0.607, p=0.000 and r= 0.931, p=0.000 respectively) (Table 4).

In unexplained male infertility group, a significant positive correlation was established between serum and seminal Vit. D levels and serum calcium level (r=0.482, p=0.007 and r=0.477, p=0.008 respectively) (Table 5).

Our study revealed in idiopathic male infertility group, a significant positive correlation between serum Vit. D level and non-progressive sperm motility (r=0.450, p=0.012). Significant negative correlations between serum and seminal Vit. D levels and serum levels of FSH and LH were found (Table 5).

Table 4: Correlations between serum and seminal Vit. D levels in different study groups.

Serum Vit. D	Seminal Vit. D		
	Control	Unexplained infertility	Idiopathic infertility
r-value	0.87	0.607	0.931
P-value	0.000*	0.000*	0.000*

*P-value ≤0.05 is significant.

Receiver Operating Characteristic (ROC) curve (Figure 1, Table 6) showed serum Vit. D level at cut off point ≤ 23.61(ng/ml) has 76.67% sensitivity and 63.33% specificity for prediction of unexplained male infertility with the Area Under Curve (AUC) was 0.699. Also, Vit. D levels at cut off point ≤21.96 (ng/ml) has 70.00% sensitivity and 76.67% specificity for prediction of idiopathic male infertility with AUC was 0.748. As well, serum levels of Vit. D at cut off point ≤22.08 (ng/ml) has 65.00% sensitivity and 76.67% specificity for prediction of male infertility either idiopathic or unexplained with AUC was 0.723.

Table 5: Correlations between serum and seminal Vit. D levels, semen parameters, male reproductive hormones and serum calcium level in different groups.

	Control group				Unexplained group				Idiopathic group			
	Serum vit. D		Seminal vit. D		Serum vit. D		Seminal vit. D		Serum vit. D		Seminal vit. D	
	r-value	p-value	r-value	P-value	r-value	p-value	r-value	p-value	r-value	p-value	r-value	p-value
Volume (ml)	-0.033	0.863	-0.029	0.88	0.286	0.126	-0.206	0.275	0.214	0.256	0.143	0.452
Sperm concentration (10^6/ ml)	-0.125	0.51	-0.108	0.569	0.053	0.783	0.017	0.929	0.148	0.436	0.01	0.958
Sperm count/ ejaculate (10^6/ ejaculate)	-0.049	0.795	-0.087	0.649	0.097	0.611	0.172	0.362	0.274	0.143	0.125	0.511
Rapid Linear Progressive motility	0.31	0.095	0.232	0.217	-0.085	0.657	-0.134	0.48	-0.173	0.147	-0.09	0.636
Slow & non-linear progressive motility	-0.269	0.151	-0.171	0.366	0.327	0.078	0.166	0.381	0.244	0.193	0.126	0.507
Progressive motility	0.043	0.82	0.05	0.794	0.11	0.561	0.024	0.899	0.073	0.703	0.076	0.689
Non-progressive motility	0.128	0.502	0.072	0.707	0.053	0.783	0.102	0.591	0.45	0.012*	0.175	0.354
Total motility	0.088	0.644	0.096	0.613	0.216	0.252	0.092	0.63	0.243	0.196	0.149	0.433
Immotile sperms	-0.106	0.577	-0.1	0.599	-0.216	0.252	-0.092	0.63	-0.243	0.196	-0.149	0.433
Sperm morphology	0.032	0.866	-0.045	0.814	0.195	0.301	0.097	0.61	0.028	0.883	-0.059	0.755
FSH (mIU/ml)	0.097	0.61	0.173	0.36	0.077	0.687	-0.191	0.311	-0.406	0.026*	-0.356	0.05*
LH (mIU/ml)	-0.095	0.619	0.163	0.388	-0.337	0.069	-0.217	0.249	-0.388	0.034*	-0.43	0.018*
Testosterone (ng/dl)	0.063	0.742	0.114	0.549	0.18	0.342	0.179	0.344	-0.16	0.4	-0.206	0.274
Calcium (mg/dl)	-0.243	0.196	-0.204	0.28	0.482	0.007*	0.477	0.008*	0.282	0.131	0.192	0.308

r = spearman correlation coefficient.
*P-value ≤0.05 is significant.

Table 6: The performance characteristics of serum Vit. D for detection of unexplained infertility, idiopathic infertility and infertility as whole.

Group	Cut-off	Sensitivity	Specificity	+PV	-PV	Accuracy	AUC
Unexplained infertility	≤ 23.61 (ng/ml)	76.67	63.33	67.6	73.1	70	0.699
Idiopathic infertility	≤ 21.96 (ng/ml)	70	76.67	75	71.9	71.67	0.748
Infertility	≤ 22.08 (ng/ml)	65	76.67	84.8	52.3	68.89	0.723

+PV: the positive predictive value, -PV: the negative predictive value, AUC: area under the curve.

Discussion

Infertility harbors massive psychosocial burdens among people and is one of the major stressors according to a study by Patel and his colleagues [25]. Pandemic Vit. D deficiency reports have extended the spectrum of extra-skeletal research on Vit. D where male reproductive function is an important area of interest [26].

In our study, we searched Vit. D level (both serum and seminal) in infertile males attending the Andrology clinic of AUH and analyzed its relation to semen parameters, serum calcium level and the levels of male reproductive hormones of men in our community.

The present study, revealed a significant decrease in both serum and seminal Vit. D in infertile males compared to healthy control group. Our results were in agreement with the results of Abbasihormozi et al. who found a significant prevalence of Vit. D deficiency and insufficiency in subfertile men suggesting that Vit. D levels may be associated with male infertility [26]. The same was found in another study performed by Alzoubi et al. who found that patients with idiopathic male infertility showed significantly lower serum levels of Vit. D at baseline when compared to controls [27].

On the contrary to our results, a previous study revealed a significant decrease in Vit. D levels in patients that had altered sperm parameters [28] Also, Zhu et al. revealed that the levels of serum Vit. D were significantly lower in asthenospermia, oligoasthenospermia and azoospermia infertile patients than those in fertile men [29].

In addition, our results revealed no significant associations among serum Vit. D and testosterone levels in our studied groups, this was in agreement with a previous study which reported that the serum level of Vit. D was not correlated with testosterone levels even after adjustment for confounding variables [30]. Also, Lerchbaum et al. [31] found no significant effect of Vit. D on androgen levels including total and free testosterone levels in their cohort of middle-aged healthy men with low baseline serum total testosterone levels. Moreover, Tirabassi et al. [32] showed no association between Vit. D and testosterone levels. Besides, heijboer et al. [33] who observed that Vit. D supplementation caused a clear increase in Vit. D levels without any statistically significant effect on testosterone levels. These results concluded the lack of significant stimulatory effects of Vit. D on testicular testosterone production. This suggests that Vit. D therapy has no clinical relevant effect on testosterone levels in males.

As regards, FSH and LH hormones, our data didn't reveal any significant correlations between serum and seminal Vit. D levels and FSH and LH serum levels in both control and unexplained male infertility groups. However, serum and seminal Vit. D levels had significant negative correlations with both FSH and LH serum levels only in the idiopathic male infertility. This was in agreement with a previous study which concluded that serum FSH and LH measures didn't differs significantly across categories of serum Vit. D concentrations among young Spanish males [34].

However, a human study done by Rehman et al. [28] revealed that

only LH levels had a non-significant negative correlation with Vit. D levels while FSH didn't. In this group of men, referred for unexplained infertility, Vit. D is not correlated with total testosterone; however it is negatively correlated with LH. As LH regulates the secretion of testosterone, these findings suggested that low Vit. D levels may suppress testicular production of testosterone that is corrected by the over production of LH.

In addition, we didn't find any significant correlations between serum Vit. D level and sperm count or concentration in all studied groups. This is in accordance with a previous study [35] that showed that there was no significant difference between semen parameters in both fertile and infertile groups with and without Vit. D deficiency. On contrary, another study performed on infertile men found a positive correlation between serum Vit. D level with both sperm concentration and progressive sperm motility which significantly improved after Vit. D treatment for 6 months [36].

In addition, a Turkish study performed on unexplained infertility showed positive correlation between Vit. D and sperm concentration [37].

Previous researches have been conducted to evaluate the association of Vit. D status and sperm motility and morphology with contrary results. Our results revealed a significant positive correlation between serum Vit. D level and non-progressive motility in idiopathic male infertility group while no associations were found between serum Vit. D level and sperm morphology in all studied groups. Azizi et al. declared that total sperms motility was positively correlated with serum concentration of 25OHD in both normozoospermic and oligoasthenoteratozoospermic men [38]. Additionally, Abbasihormozi et al. [26] revealed that Vit. D levels showed no correlation with sperm parameters and hormone profiles in normospermic men, however, sperm motility and calcium were positively correlated with Vit. D in oligoasthenoteratozoospermic group. Another study, showed a statistically significant correlation between base line serum Vit. D level and all semen parameters. After treatment with vitamin D, there was a statistically significant improvement only in sperm progressive forward motility and total sperm motility after Vit. D restoration [28]. Similarly, a previous study reported that the incidence of hypovitaminosis D among men seeking fertility therapy is similar to the national average and oral Vit. D treatment improves sperm motility among these males [39].

Conclusion

The presence of significant decrease of both serum and seminal Vit. D levels in infertile males compared to controls and the significant positive correlation between serum Vit. D level and non-progressive sperm motility; suggesting the role of Vit. D in male fertility. However, there was no correlations between serum levels of Vit. D and male reproductive hormones; indicating that Vit. D effect in male fertility may not be mediated through the male reproductive hormones.

References

1. World Health Organization. Infertility definitions and terminology. 2018.
2. Zegers-Hochschild F, Adamson GD, Dyer S, Racowsky C, De Mouzon J, Sokol R, et al. The International Glossary on Infertility and Fertility Care. Fertil Steril. 2017; 108: 393-406.
3. World Health Organization. WHO laboratory manual for the examination and processing of human semen. 2010.
4. Agarwal A, Mulgund A, Hamada A, Chyatte MR. A unique view on male infertility around the globe. Reprod Biol Endocrinol. 2015; 13: 37.
5. Ring JD, Lwin AA, Köhler TS. Current medical management of endocrine-related male infertility. Asian J Androl. 2016; 18: 357-363.
6. Lalitha C, Sayee R, Jeyanthi K, Shubha R. Hormonal factors associated with hypogonadism and infertility in males-chromosomal abnormality. J Dent Med Sci. 2013; 10: 71-75.
7. Al-Faisal A. Hormonal disturbances among the infertile men in Baghdad-Iraq. J Med Tech Assoc Thailand. 2010; 38: 3060-3066.
8. Al-Daghistani H, Abdel-Dayem M. Hyperprolactinemia and Hypergonadotropins in Infertile Males with Severe Oligospermia and Azoospermia. The Internet J Endocrinol. 2006; 3.
9. Singh P, Singh M, Cugati G, Singh A. Hyperprolactinemia. An often missed cause of male infertility. J Hum Reprod Sci. 2011; 4: 102-103.
10. Jungwirth A, Diemer T, Kopa Z, Krausz C, Minhas S, Tournaye H. EAU guidelines on male infertility. Arnhem: European Association of Urology. 2018.
11. Agarwal A, Parekh N, Panner Selvam MK, Henkel R, Shah R, Homa ST, et al. Male Oxidative Stress Infertility (MOSI): Proposed terminology and clinical practice guidelines for management of idiopathic male infertility. World J. Men's Health. 2019; 37: 296-312.
12. Doshi SB, Sharma RK, Agarwal A. Oxidative stress in unexplained male infertility. In Unexplained Infertility: Pathophysiology, Evaluation and Treatment. Eds.; Springer: New York, NY, USA. 2015; 81-89.
13. Carlberg C. Nutrigenomics of vitamin D. Nutrients. 2019; 3: E676.
14. Charoenngam N, Shirvani A, Holick MF. Vitamin D for skeletal and non-skeletal health: What we should know. J Clin Orthop Trauma. 2019; 10: 1082-1093.
15. Mutchie TR, Yu OB, Di Milo ES, Arnold LA. Alternative binding sites at the vitamin D receptor and their ligands. Mol Cell Endocrinol. 2019; 485: 1-8.
16. Taglianetti S, De Rocco PM, Ghezzi M, Foresta C. Vitamin-D Deficiency and Male Reproduction. Ann Nutr Disord and Ther. 2016; 3: 2381-8891.
17. Yao X, El-Samahy MA, Yang H, et al. "Age-associated expression of vitamin D receptor and vitamin D-metabolizing enzymes in the male reproductive tract and sperm of Hu sheep. Animal Reproduction Science. 2019; 190: 27-38.
18. Bollehuus HL, Rehfeld A, Neergaard RD, Nielsen JE, Iversen LH, Boisen IM, et al. Selection of high-quality spermatozoa may be promoted by activated vitamin D in the woman. J Clin Endocrinol Metab. 2017; 102: 950-961.
19. Aquila S, Guido C, Middea E, Perrotta I, Bruno R, Pellegrino M, et al. Human male gamete endocrinology: 1alpha, 25-dihydroxyvitamin D3 (1,25(OH)2D3) regulates different aspects of human sperm biology and metabolism. J Reproductive Biology Endocrinology. 2009; 7: 140.
20. Moghadam MT, Hosseini G, Absalan F, Tabar MH, Nikbakht R. Effects of Vitamin D on Apoptosis and Quality of Sperm in Asthenozoospermia JBRA Assisted Reproduction. 2020; 24: 316-323.
21. De Angelis C, Galdiero M, Pivonello C, Garifalos F, Menafra D, Cariati F, et al. The role of vitamin D in male fertility: a focus on the testis. Rev Endocr Metab Disord. 2017; 18: 285-305.
22. Blomberg Jensen M, Bjerrum PJ, Jessen TE, Nielsen JE, Joensen UN, Olesen IA, et al. Vitamin D is positively associated with sperm motility and increases intracellular calcium in human spermatozoa. Human Reproduction. 2011; 26: 1307-1317.
23. Jueraitetibaike K, Ding Z, Wang DD, Peng L. P, Jing J, Chen L, et al. The effect of vitamin D on sperm motility and the underlying mechanism. Asian J Androl. 2019; 21: 400-407.
24. Pfotenhauer KM, Shubrook JH. Vitamin D deficiency, its role in health and disease, and current supplementation recommendations. J Am Osteopath Assoc. 2017; 117: 301-305.

25. Patel A, Sharma PSVN, Narayan P, Binu VS, Dinesh N, Pai PJ. Prevalence and predictors of infertility-specific stress in women diagnosed with primary infertility: A clinic-based study. Journal of human reproductive sciences. 2016; 9: 28-34.

26. Abbasihormozi S, Kouhkan A, Alizadeh AR, Shahverdi AH, Nasr-Esfahani MH, Sadighi Gilani MA, et al. Association of vitamin D status with semen quality and reproductive hormones in Iranian subfertile men. Andrology. 2017; 5: 113-118.

27. Alzoubi A, Mahdi H, Al Bashir S, Halalsheh O, Al Ebbini M, Alzarir M, et al. Normalization of serum vitamin d improves semen motility parameters in patients with idiopathic male infertility. Acta endocrinologica (Bucharest, Romania: 2005). 2017; 13: 180-187.

28. Rehman R, Lalani S, Baig M, Nizami I, Rana Z, Gazzaz ZJ. Association Between Vitamin D, Reproductive Hormones and Sperm Parameters in Infertile Male Subjects. Frontiers in endocrinology. 2018; 9: 607-607.

29. Zhu C, Xu QF, Li SX, Wei YC, Zhu G C, Yang C, et al. Investigation of serum vitamin D levels in Chinese infertile men. Andrologia. 2016; 48: 1261-1266.

30. Sim MY, Kim SH, Kim KM. Seasonal Variations and Correlations between Vitamin D and Total Testosterone Levels. Korean J Fam Med. 2017; 38: 270-275.

31. Lerchbaumm E, Trummer C, Theiler-Schwetz V, Kollmann M, Wölfler M, Heijboer AC, et al. Effects of vitamin D supplementation on androgens in men with low testosterone levels: a randomized controlled trial. Eur J Nutr. 2019; 58: 3135-3146.

32. Tirabassi G, Cutini M, Muscogiuri G, Delli Muti N, Corona G, Galdiero M, et al. Association between vitamin D and sperm parameters: Clinical evidence. Endocrine. 2017; 58: 194-198.

33. Heijboer AC, Oosterwerff M, Schroten NF, Eekhoff EM, Chel VG, de Boer RA, et al. Vitamin D supplementation and testosterone concentrations in male human subjects. Clinical endocrinology. 2015; 83: 105-110.

34. Rudnicka A, Adoamnei E. Vitamin D status is not associated with reproductive parameters in young Spanish men. 2020; 8: 323-331.

35. Zahraa S, Batool Shahraki M, Alireza S. Comparison of Vitamin D Levels in Fertile and Infertile Men. Maedica (Bucur). 2020; 15: 96-98.

36. Wadhwa L, Priyadarshini S, Fauzdar A, Wadhwa S. N, Arora S. Impact of Vitamin D Supplementation on Semen Quality in Vitamin D-Deficient Infertile Males with Oligoasthenozoospermia. J Obstet Gynaecol India. 2020; 70: 44-49.

37. Ozdemir E, Tokmak A, Erkilinc S, Yakut HI, Erkaya S, Yilmaz N. Association between vitamin D levels and semen parameters in infertile males. Journal of Clinical and Analytical Medicine. 2015; 6: 765-769.

38. Azizi E, Naji M, Shabani-Nashtaei M, Aligholi A, Najafi A, Amidi F. Association of serum content of 25-hydroxy vitamin D with semen quality in normozoospermic and oligoasthenoteratozoospermic men. International Journal of Reproductive Bio Medicine. 2018; 16: 689.

39. Waud K, Bocca S. Role of vitamin D deficiency in male fertility. Fertility and Sterility. 2015; 103: 39.