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RELATIONSHIP BETWEEN VITAMIN D AND DISEASE ACTIVITY IN SOME RHEUMATIC DISEASES

Mohamed Hashem Ibrahim
Rheumatology, Physical Medicine and Rehabilitation Department, Makka, KSA

Mohamad Sayyed Bakheet

Biochemistry Department, Al-Azhar Faculty of Medicine, Assiut, Egypt

Khaled A Abdel-Sater

Physiology Department, Al-Azhar Faculty of Medicine, Assiut, Egypt

Maher Abdel shakoor

Physiology Department, Al-Azhar Faculty of Medicine, Assiut, Egypt

ABSTRACT: *Vitamin D is recognized as an important immuno-modulatory factor involved in autoimmune rheumatic diseases. The effect of vitamin D on the immune system is an enhancement of innate immunity coupled with multifaceted regulation of adaptive immunity. 1,25(OH)2D3, the biologically active metabolite of Vitamin D, not only regulates bone and calcium metabolism but also exerts immuno-modulation via the nuclear vitamin D receptors (VDR) expressed in antigen-presenting cells and activated T/B cells . In the current study 120 volunteers divided into two groups: patients group and control group. Patients group included 100 rheumatic patients, including 30 Rheumatoid arthritis (RA) patients , 20 SLE patients ,30 osteoarthritis patients(OA) , 10 Behçet's disease patients , 10 ankylosing spondylitis (AS)patients. The Control group included 20 healthy volunteers. The age of the patients ranged between 16 - 65 years. The disease duration ranged from 1 to 20 years.*
The mean value of vitamin D serum level (ng/mL) was found to be low in RA group (mean ± SD)(13.47 ± 8.17) in comparison to control group(26.61 ± 6.44). There was no statistically significant difference in the mean value of vitamin D level between the RA active group and inactive group. There was vitamin D deficiency in SLE patients=(19.32 ±10.67), the difference in the mean value of vitamin D serum level between the SLE active group and inactive group was found to be statistically insignificant. The difference in the mean value of vitamin D serum level between the OA group (22.95 ±9.30) and control group(26.61 ± 6.44) was found to be statistically insignificant, as p-value of 0.178.In the O.A. group, the difference in the mean value of vitamin D serum level between the O.A. active group and inactive group was found to be statistically insignificant. The vitamin D serum levels were lower in the AS group (17.81 ±8.11) than the control group. The difference in the mean value of vitamin D level between the A.S. group and control group was found to be statistically significant. As regard the comparison of vitamin D serum level between the active A.S. group and the inactive A.S. group, the difference was found to be statistically insignificant, as p-value of 0.17. The mean value of vitamin D level in the Behçet's disease group was significantly lower (17.64 ±8.79) in comparison to its level in the control group.There was no statistical significance difference in the mean value of vitamin D serum level in the active and the inactive Behcet disease groups.

KEYWORDS: Vitamin D, immune system, autoimmune diseases, systemic lupus erythematosus, rheumatoid arthritis

INTRODUCTION

Vitamin D is one of the fat soluble vitamins which derived from Cholecalciferol (7-Dehydrocholesterol, Pro-vitamin D3) in human and from ergosterol (Pro-vitamin D2) in yeast and plants, both forms are bioactive. The main source of vitamin D is de novo synthesis in the skin. Vitamin D_3 is produced in the skin of vertebrates after exposure to ultraviolet B light from the sun (Koyyalamudi et al., 2009).

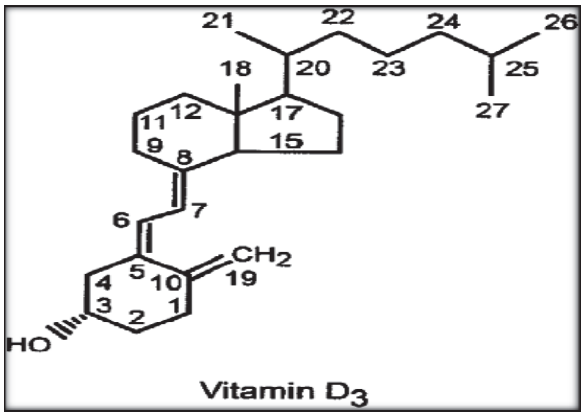

Fig. (1) Cholecalciferol (D3) (Adams, and Hewison, 2010).

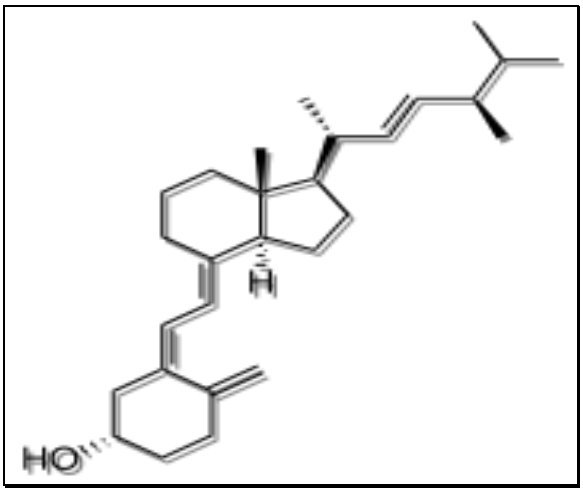

Fig. (2) Ergocalciferol (D2) (Joshi,et al., 2010).

Vitamin D3 (cholecalciferol) is hydroxylated in the liver at position 25 forming 25-hydroxycholecalciferol (calcidiol). This reaction is catalyzed by the microsomal enzyme vitamin D 25-hydroxylase, which is produced by hepatocytes. Once made the product is stored in the hepatocytes until it is needed and then can be released into the plasma where it will be bound to an α-globulin (Cheng et al., 2004).

Calcidiol is then converted in the kidneys (by the enzyme 25(OH)D-1α-hydroxylase) into calcitriol (1,25-(OH)2D3), a secosteroid hormone that is the active form of vitamin D. It can also be converted into 24-hydroxycalcidiol in the kidneys via 24-hydroxylation. This product

is a potent ligand of the vitamin D receptor (VDR) which mediates most of the physiological actions of the vitamin. (Arnson et al., 2007).

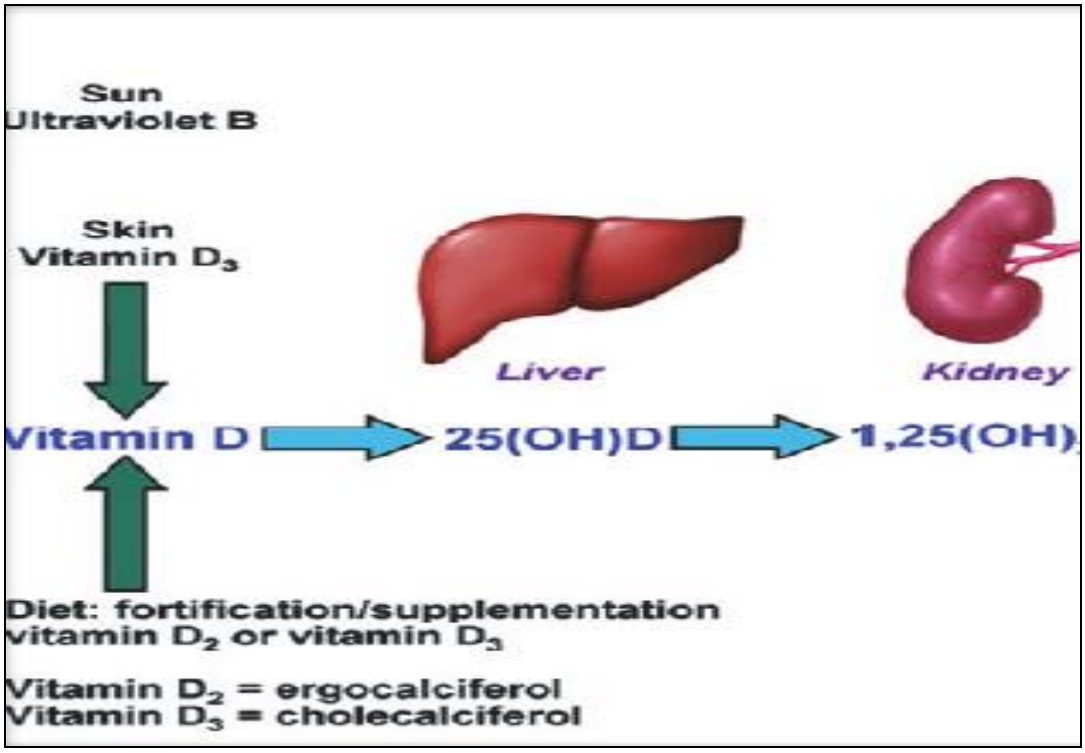

Fig. (3) Mechanism of synthesis of vitamin D; 1, 25dihydroxyvitamin D (Cheng et al., 2004).

The binding of calcitriol to the VDR allows the VDR to act as a transcription factor that modulates the gene expression of transport proteins (such as TRPV6 and calbindin), which are involved in calcium absorption in the intestine. The vitamin D receptor belongs to the nuclear receptor superfamily of steroid/thyroid hormone receptors, and VDRs are expressed by cells in most organs, including the brain, heart, skin, gonads, prostate, and breast. VDR activation in the intestine, bone, kidney, and parathyroid gland cells leads to the maintenance of calcium and phosphorus levels in the blood (with the assistance of parathyroid hormone and calcitonin) and to the maintenance of bone content. (Holick, 2004).

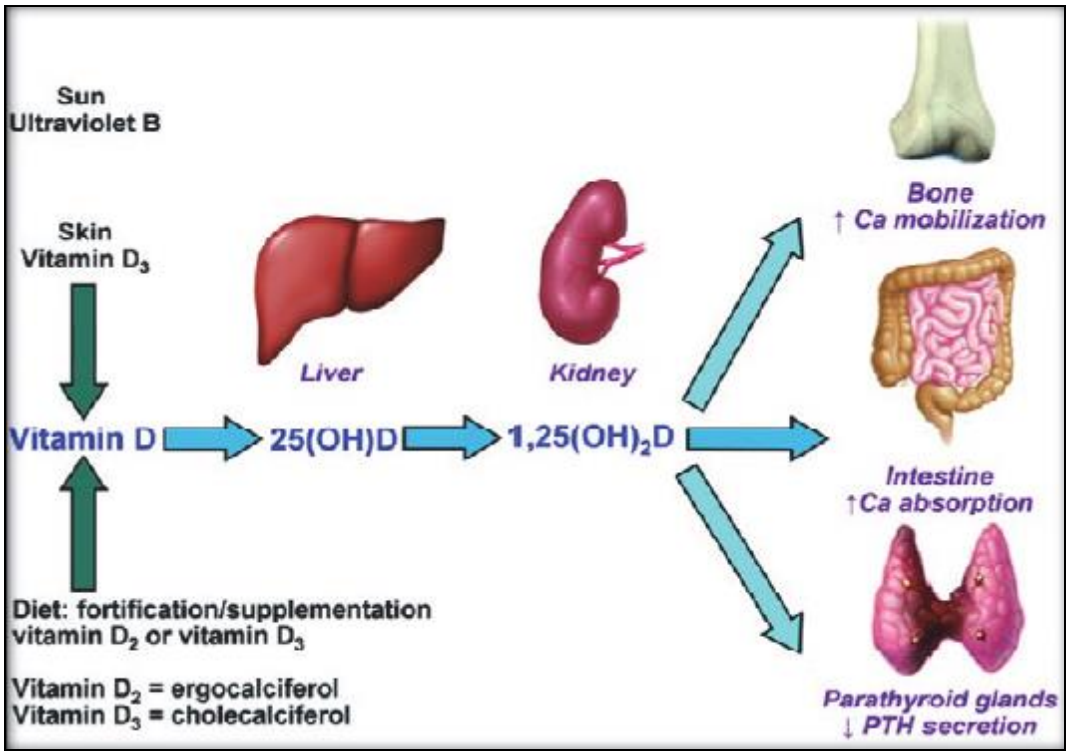

Fig. (4) Role of Synthesis and Metabolism of Vitamin D in the regulation of Calcium and Bone mineralization (Holick et al., 2007).

Lack of vitamin D activity leads to reduced intestinal absorption of calcium and phosphorus. Early in hypovitaminosis D, hypophosphatemia is more marked than hypocalcemia. With persistent hypovitaminosis D, hypocalcemia causes a secondary hyperparathyroidism that leads to phosphaturia, demineralization of bones, and without treatment, to osteomalacia in adults and rickets in children. Glucocorticoids, when used chronically in high doses, inhibit the intestinal vitamin D dependent calcium absorption and therefore cause osteomalacia. Sub clinical vitamin D deficiency (or vitamin D insufficiency) is extremely common and may contribute to the development of osteoporosis. Vitamin D stores decline with age, especially in the winter. Controlled trials have demonstrated that vitamin D and calcium supplementation can reduce the risk of falls and fractures in the elderly (Misra et al., 2008).

1,25-dihydroxyvitamin D3, the biologically active metabolite of Vitamin D3, not only regulates bone and calcium metabolism but also exerts immunomodulation via the nuclear VDR expressed in antigen-presenting cells and activated T/B cells (Van Etten and Mathieu 2005).

The effect of vitamin D on the immune system is an enhancement of innate immunity coupled with multifaceted regulation of adaptive immunity (Adorini and Penna 2008).

The discovery of the vitamin D receptors (VDR) in the cells of the immune system and the fact that several of these cells produce vitamin D hormone suggested that it could have immunoregulatory properties (Sigmundsdottir et al., 2007).

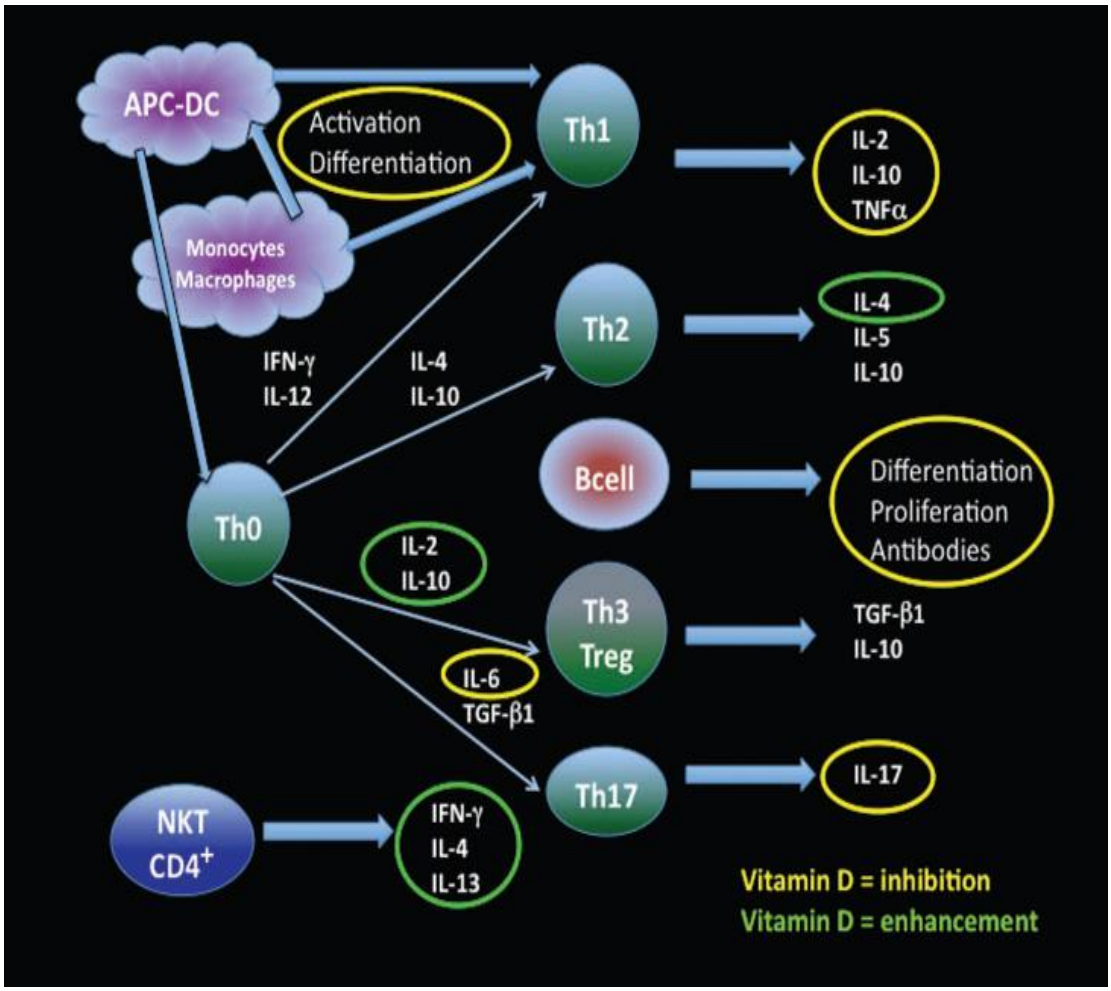

Fig. (5) Mechanisms involved in vitamin D modulation of the immune responses. Dendritic cells (DCs) are primary targets for the immunomodulatory activity of 1,25(OH)2D3, as indicated by inhibited DC differentiation and maturation, together with inhibition of differentiation of monocyte precursors into immature DCs. 1,25(OH)2D3 suppresses Th1 (and Th17) driven cytokine responses, induces Treg cells, induces IL-4 production (Th2) and enhances NKT-cell function. Differentiation and maturation of B cells is also inhibited. Th are CD4þ helper cell subsets (Th1, Th2, Th3-Treg, Th17) originating from naı¨ve T cell (Th0). Thin arrows (left) indicate cytokines that induce differentiation of Th0 cells and thicker arrows (right) indicate cytokines produced by activated Th cell subsets. All T cells that have been tested express the VDR. B cells and NKT cells are also reported.The yellow circles indicate the cytokines/activities inhibited by vitamin D. On the contrary, the green circles indicate the cytokines enhanced by vitamin D. (Sigmundsdottir et al., 2007).

Table (1) Action of Vitamin D in immune system

Target cell population	Actions mediated by 1. 25(OH)2 D3
antigen-presenting cell APCs (monocytes, macrophages, dendritic cells)	inhibits the expression of class II major histo-compatibility complex MHC molecules, inhibits the expression of costimulating molecules (CD40, CD80, and CD86) and other maturation inducing proteins (CD1a, CD83), increases chemotaxis and phagocytosis of monocytes and cytotoxicity against tumor cells and bacteria, inhibits the maturation of dendritic cells, induces tolerogenic dendritic cells capable of inducing Treg cells, inhibits the release of interleukin IL-12 p70 and inhibits pro-inflammatory cytokines (IL-1 and tumor necrosis factor TNF) by monocytes and macrophages.
T lymphocytes	inhibits T cell proliferation, secretion of cytokines, and progression of the cellular cycle from G1a to G1b, increases the production of IL-4, IL-5, IL-10 inhibits IL-12, ɤ gmma interferon INF-γ, and IL-2 inhibits activation of antigen specific T lymphocytes inhibits the expression of Fas ligand (FasL) by activated T lymphocytes
B cells	Expresses VDR Suppresses IgE secretion
natural killer NK cells	inhibits INF-γ

(Cantorna et al., 2008).

However, vitamin D insufficiency is emerging as a clinical problem of global proportions and epidemiology has linked vitamin D status with autoimmune disease susceptibility and severity, epidemiological evidence indicates a significant association between vitamin D deficiency and an increased incidence of a variety of autoimmune rheumatic diseases such as rheumatoid arthritis (RA) and SLE (Adorini and Penna 2008) .

Observational studies in humans suggest an association between vitamin D deficiency and many rheumatological and non-rheumatological disorders, listed in Table 2

Table (2) Disorders that have been linked to 1. 25(OH) 2 D3

Rheumatological disorders	Non Rheumatological disorders
1. Rheumatoid Arthritis "RA". 2. Undifferentiated Connective tissue UCTD. 3. Systemic lupus erythematosus SLE. 4. Scleroderma. 5. Ankylosing spondylitis"AS" . 6. Behcet's disease. 7. Psoriasis. 8. Fibromylgia .	1. Multiple Sclerosis "MS". 2. Insulin dependent Diabetes Mellitus "IDDM". 3. Allergic asthma in children. 4. Allergic rhinitis. 5. Grave's disease.

(Cutolo and Otsa 2008).

Low serum levels of vitamin D3 might be partially related, among other factors, to prolonged daily darkness (reduced activation of the pre-vitamin D by the ultraviolet B sunlight),

different genetic background (i.e. vitamin D receptor polymorphism) and nutritional factors, and explain to the latitute-related prevalence of autoimmune diseases such as RA, by considering the potential immunosuppressive roles of vitamin D.

Treatment of vitamin D deficiency could be particularly important in SLE patients due to concomitant insults on their tissues such as bone, and in view of the discovered immunomodulatory effects exerted by vitamin D (Cutolo, 2008).

Low sun exposure and reduced body mass index (BMI) are well established risk factors for vitamin D deficiency in RA patients. (Rossini et al., 2010).

Few studies have examined dietary or nutritional intake prior to RA onset, and none have assessed the association of vitamin D with disease onset. Merlino et al., in 2004 found that: greater intake of total daily vitamin D was inversely associated with risk of RA. Inverse associations were apparent for both dietary and supplemental vitamin D.

Several studies have demonstrated a higher prevalence of vitamin D deficiency in SLE patients when compared to individuals with other rheumatologic diseases and healthy controls (Borba et al., 2009).

The inflammatory activity in ankylosing spondylitis (AS) itself plays a major role in the pathophysiology of bone loss; this may be mediated in AS by substances regulating both the inflammatory process and bone turnover. High levels of pro-inflammatory cytokines such as interleukin-1 and tumor necrosis factor α (TNFα) are thought to play a major role in chronic inflammation and act on osteoblasts and osteoclasts (Lange et al., 2005).

Osteoporosis is frequent in AS and high disease activity which assessed by Bath Ankylosing Spondylitis Disease Activity Index (BASDAI) is associated with an alteration in vitamin D metabolites and increased levels of bone resorption (Braun-Moscovici et al., 2008) .

Zold et al., 2008 demonstrated the presence of a seasonal variation in the levels of 1. 25(OH) 2 D3in patients with UCTD and that those levels were lower in this population than in the control population. In the same study, 21.7% of patients with UCTD and vitamin D deficiency developed established connective tissue disease (especially RA, SLE, Sjogren's syndrome, and mixed connective tissue disease); their mean 1. 25(OH) 2 D3 was lower than that of patients who remained with undifferentiated disease, 14. 7± 6. 45 ng/mL vs 33. 0 ± 13. 4 ng/mL, P =0. 0001 respectively.

Prospective studies available for the 4 major autoimmune diseases :RA, SLE, MS, and type 1 DM, have demonstrated the beneficial effects of vitamin D supplementation in modulating the components of the immune system responsible for the inflammation, such as the expression of cytokines, growth factors, nitrous oxide, and metalloproteinase (Marques et al., 2010).

The aim of this work is to estimate the level of 1, 25(OH) 2 D3 in different rheumatic diseases to find the relation between 1, 25(OH) 2 D3 level and rheumatic diseases and to establish its relation to the rheumatic diseases activity and severity.

SUBJECTS AND METHODS

This study was done on 100 patient selected from the outpatient clinic of rheumatology department fuculty of medicin Alazher university Assuit branch as patients group, their age ranged between 16 years - 65 years old, The disease duration ranged from 1 to 20 years. The following patients were excluded from the study: patients who had parathyroid disorder, patients who had renal disorder, patients who had hepatic disorder, patients who had gastrointestinal and metabolic disorders, patients who had diabetes and patients who received vitamin D supplementation.

The study also included 20 healthy volunteers as control group who were match the patients group in age and socio-economic status.

The study has been approved by the relevant research and ethics committee and after informed consent for each of patients and control groups.

Patients group subdivided into: 30 patients suffering of R.A., 30 patients suffering of O.A., 20 patients suffering of SLE. , 10 patients suffering of Behcets disease and 10 patients suffering of ankylosing spondylitis.

The following items were done for each of patients and control groups:-

- Medical history, general clinical examination, body joint examination to determine joint tenderness, arthritis, tenosynovitis, deformity or functional limitation of the affected joints, muscular examination for atrophy, tenderness and weakness.
- Venous blood samples were taken for determination of complete blood count using automated cell counter, E.S.R using westergren tubes method, serum 1. 25(OH) 2 D3 level, serum calcium (total and ionized),serum phosphorus, serum Parathormone, blood urea and serum creatinine ,liver function tests (AST&ALT) ,rheumatoid factor using latex agglutination test, ANA &Anti double stranded DNA and Anti Sm ab, fasting and post prandial blood glucose level and complete urine analysis by microscopic examination.
- Radiology: - Plain X-ray of hands and feet (postero-anterior view].

  - Plain X-ray of other affected joints.
- The disease activity in different target groups was assessed using Disease activity score 28 (DAS28) in R.A. patients, SLE Disease Activity Index (SLEDAI) in SLE patients, Bath Ankylosing Spondylitis Disease Activity Index (BASDAI) in A.S. patients, The Western Ontario and McMaster Universities Arthritis Index (WOMAC osteoarthritis index) in O.A. patients and American College of Rheumatology (ACR) criteria in Behcet patients.

Assessment of 1. 25(OH) 2 D3 sufficiency by in vitro quantitative determination of 1. 25(OH) 2 D3 in human serum by using the electrochemiluminescence immunoassay ECLIA (Leino et al., 2008).

ECLIA is a highly innovative technology that offers distinct advantages over other detection techniques including: extremely stable non-isotopic label allows liquid reagent convenience,

enhanced sensitivity in combination with short incubation times, means high quality assays and fast result turnaround, large measuring range of five orders of magnitude minimizes dilutions and repeats, reducing handling time and reagent costs and applicable for the detection of all analytes providing a solid platform for menu expansion (Weir, D M - Elecsys 2010).

ECLIA is based on competition principle. Total duration of assay: 18 minutes.

1st incubation: 1. 25(OH) 2 D3 in the sample (35µL) competes with the biotin labeled vitamin D in the complex contained in R2 (biotin-vitamin D/polyclonal 1. 25(OH) 2 D3 -specific ruthenium labeled antibody). The remaining amount of the complex (biotin-vitamin D/polyclonal 1. 25 (OH) 2 D3 -specific ruthenium labeled antibody) is dependent upon the analyte concentration in the sample.

2nd incubation: After addition of streptavidin-coated microparticles the complex becomes bound to the solid phase via interaction of biotin and strepavidin. The reaction mixture is aspirated into the measuring cell where the microparticles are magnetically captured onto the surface of the electrode. Unbound substances are then removed with ProCell. Application of a voltage to the electrode then induces chemiluminescent emission which is measured by a photomultiplier. Results are determined via a calibration curve which is instrument-specifically generated by 2-point calibration and a master curve provided via the reagent barcode (Roth et al., 2008).

The analyzer automatically calculates the analyte concentration of each sample (either in ng/mL or nmol/L).

Conversion factors: nmol/l x 0.40 = ng/mL

ng/mL x 2.50= nmol/L

Data were expressed as Mean ± standard deviation , Comparisons were performed for normal distributed data using T. test for independent groups, P. value considered insignificant if it >0.05 ,significant if it ≤0.05, highly significant if it ≤0.001, the statistical analysis were done using SPSS V11.0 program.

RESULTS

Table (3) Demographic data and duration of illness in the patients group:

	R.A. group *(n = 30)* Mean ± S.D	**SLE group *(n =20)* Mean ± S.D**	**O.A Group *(n = 30)* Mean ± S.D**	**Behcet disease group *(n = 10)* Mean ± S.D**	**A.S group *(n = 10)* Mean ± S.D**	**P-value**	**Statistical Significance**
Age (years)	42.30± 12.97	37.65 ± 12.37	62.27± 10.35	40.30 ± 7.83	41.90 ± 11.70	0.000	Sig
Age of onset (years)	32.57 ± 8.23	27.35 ± 8.80	49.30 ± 15.67	26.70 ± 5.03	33.20 ± 8.28	0.000	Sig
Duration of illness(years)	9.40 ± 6.41	10.30 ± 5.91	12.93 ± 7.32	11.60 ± 4.48	8.70 ± 4.69	0.975	N.S.

Table (4) Statistical comparison of vitamin D serum level, ESR and CRP between the target R.A. patients and control group:

Characteristic	R.A. group *(n = 30)* Mean ± S.D	Control group *(n = 20)* Mean ± S.D	P-value	Statistical Significance
Vitamin D serum level in ng/mL	13.47± 8.17	26.61± 6.44	0.000	Sig.
CRP mg/dL	4.34±3.70	0.80± 0.00	0.000	Sig.
ESR mm/hr.	53.00± 24.17	24.05± 10.38	0.000	Sig.

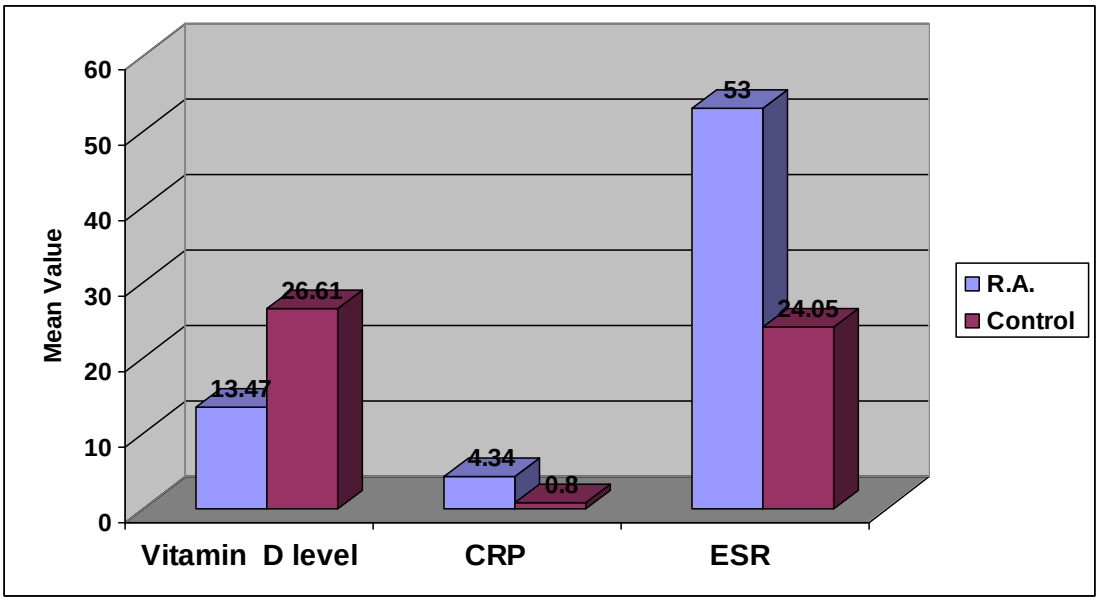

Fig. (6) Statistical comparison of vitamin D serum level, ESR and CRP between the target R.A. patients and control group.

Table (5) Statistical comparison of vitamin D serum level, ESR and CRP between the active R.A. group and the inactive R.A. group:

Characteristic	Active R.A. group *(n = 15)* Mean ± S.D	Inactive R.A. group *(n = 15)* Mean ± S.D	P-value	Statistical Significance
Vitamin D serum level in ng/mL	13.55± 6.76	13.40± 9.46	0.574	N.S
CRP mg/dL	7.93± 2.05	1.21± .60	0.000	Sig.
ESR mm/hr.	75.64± 13.55	33.19± 8.53	0.000	Sig.

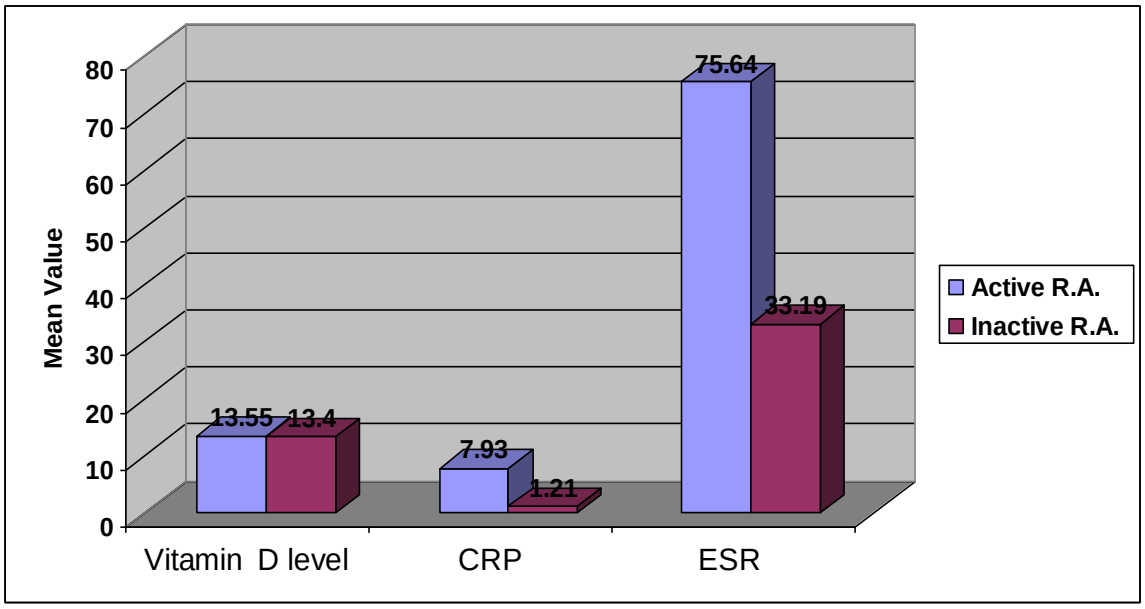

Fig. (7) Statistical comparison of vitamin D serum level, ESR and CRP between the active R.A. group and the inactive R.A. group.

Table (6) Statistical comparison of vitamin D serum level, ESR and CRP between the target SLE patients and control group:

Characteristic	SLE group *(n = 20)* Mean ± S.D	Control group *(n = 20)* Mean ± S.D	P-value	Statistical Significance
Vitamin D serum level in ng/mL	19.32± 10.67	26.61± 6.44	0.019	Sig.
CRP mg/dL	3.60±3.04	0.80± 0.00	0.000	Sig.
ESR mm/hr.	51.30± 19.15	24.05± 10.38	0.000	Sig.

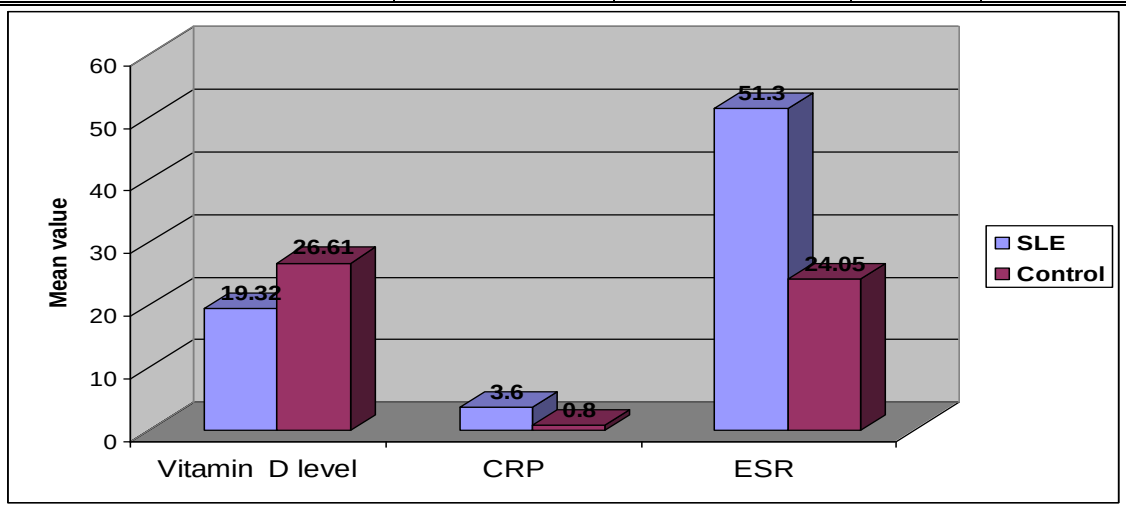

Fig. (8) Statistical comparison of vitamin D serum level, ESR and CRP between the target SLE patients and control group.

Table (7) Statistical comparison of vitamin D serum level ,ESR and CRP between the active SLE group and the inactive SLE group:

Characteristic	Active SLE group *(n = 5)* Mean ± S.D	Inactive SLE group *(n = 5-)* Mean ± S.D	P-value	Statistical Significance
Vitamin D serum level in ng/mL	18.04± 13.37	20.36± 8.41	0.470	N.S.
CRP mg/dL	6.49± 2.13	1.24± 0.55	0.000	Sig.
ESR mm/hr.	66.00± 17.17	39.27± 10.35	0.003	Sig.

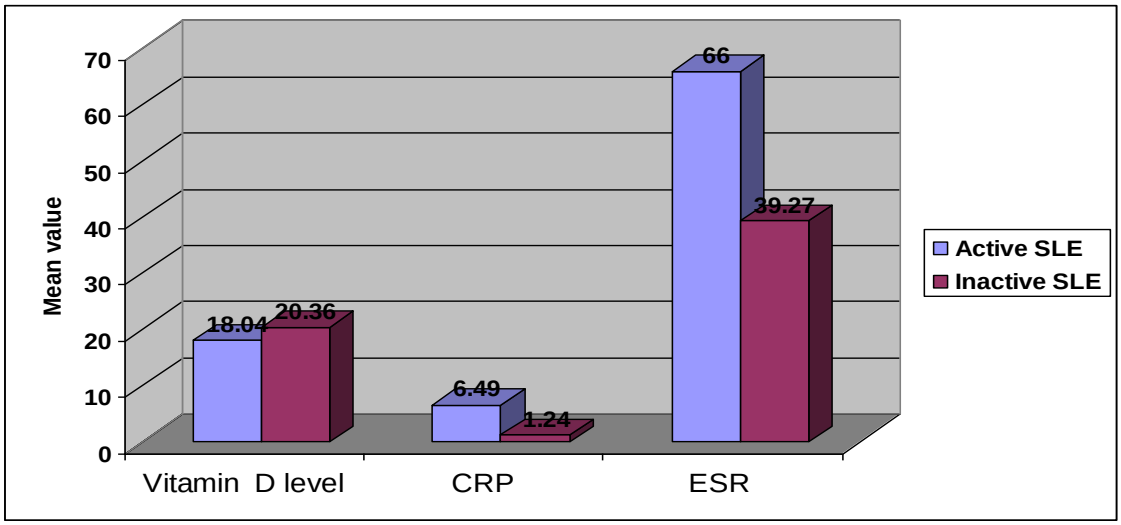

Fig. (9) Statistical comparison of vitamin D serum level, ESR and CRP between the active SLE group and the inactive SLE group.

Table (8) Statistical comparison of vitamin D serum level, ESR and CRP between the target 0.A. patients and control group:

Characteristic	0.A. group *(n = 30)* Mean ± S.D	Control group *(n = 20)* Mean ± S.D	P-value	Statistical Significance
Vitamin D serum level in ng/mL	22.95± 9.30	26.61± 6.44	0.178	N.S.
CRP mg/dL	1.25±0.80	0.80± 0.00	0.001	Sig.
ESR mm/hr.	35.00±13.72	24.05± 10.38	0.006	Sig.

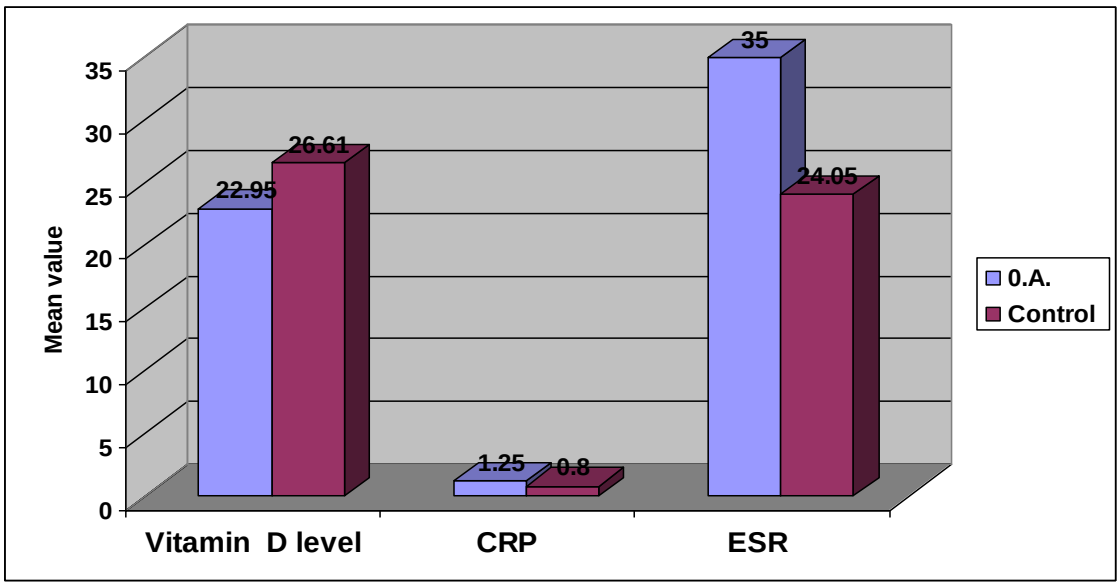

Fig. (10) Statistical comparison of vitamin D serum level, ESR and CRP between the target 0.A. patients and control group.

Table (9) Statistical comparison of vitamin D serum level, ESR and CRP between the active O.A. and the inactive O.A. group:

Characteristic	Active O.A group *(n = 15)* Mean ± S.D	Inactive O.A. group *(n = 15)* Mean ± S.D	P-value	Statistical Significance
Vitamin D serum level in ng/mL	21.73± 9.57	24.02± 9.24	0.430	N.S.
CRP mg/dL	1.60± 0.70	0.95± 0.32	0.003	Sig.
ESR mm/hr.	43.93± 10.45	27.19± 11.39	0.001	Sig.

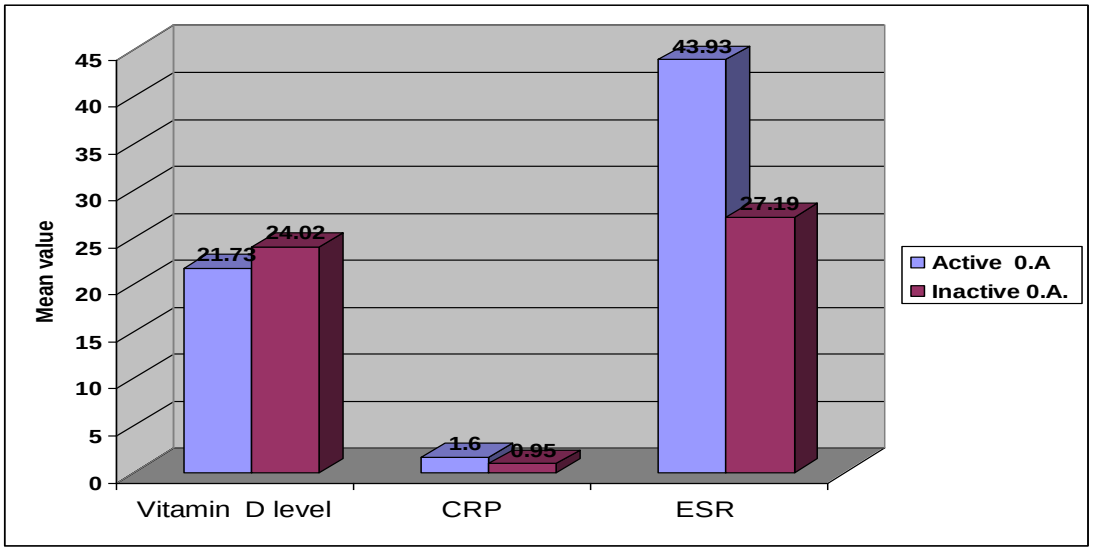

Fig. (11) Statistical comparison of vitamin D serum level, ESR and CRP between the active O.A. and the inactive O.A. group.

Table (10) Statistical comparison of vitamin D serum level, ESR and CRP between the target Behcet disease patients and control group:

Characteristic	Behcet disease group *(n = 10)* Mean ± S.D	Control group *(n = 20)* Mean ± S.D	P-value	Statistical Significance
Vitamin D serum level in ng/mL	17.64± 8.79	26.61± 6.44	0.006	Sig.
CRP mg/dL	1.85±1.76	0.80± 0.00	0.000	Sig.
ESR mm/hr.	38.70±18.51	24.05± 10.38	0.041	Sig.

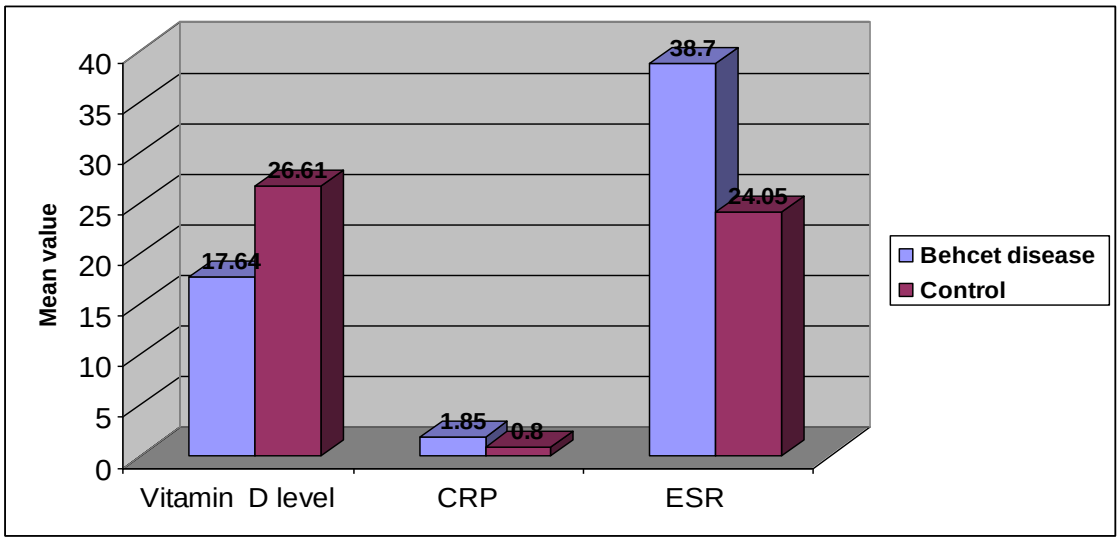

Fig. (12) Statistical comparison of vitamin D serum level, ESR and CRP between the target Behcet disease patients and control group.

Table (11) Statistical comparison of vitamin D serum level, ESR and CRP between the active Behcet disease and the inactive Behcet disease group:

Characteristic	Active Behcet disease group *(n = 5)* Mean ± S.D	Inactive Behcet disease group *(n = 5-)* Mean ± S.D	P-value	Statistical Significance
Vitamin D serum level in ng/mL	14.74± 7.70	20.54± 9.66	0.465	N.S
CRP mg/dL	2.72± 2.23	0.98± 0.35	0.081	N.S
ESR mm/hr.	53.80± 11.03	23.60± 8.88	0.009	Sig.

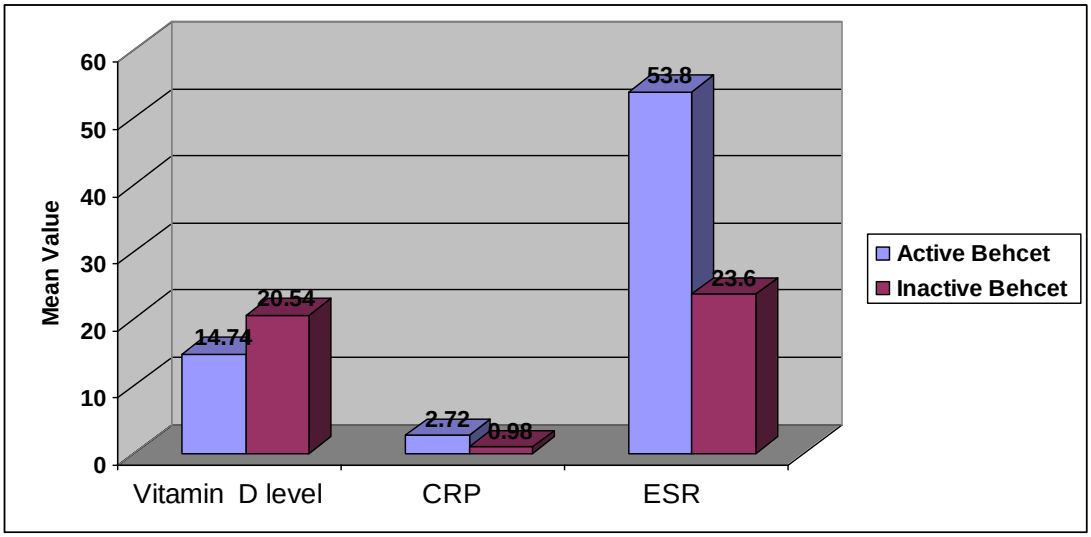

Fig. (13) Statistical comparison of vitamin D serum level, ESR and CRP between the active Behcet disease and the inactive Behcet disease group.

Table (12) Statistical comparison of vitamin D serum level, ESR and CRP between the target A.S patients and control group:

Characteristic	A.S group *(n = 10)* Mean ± S.D	Control group *(n = 20)* Mean ± S.D	P-value	Statistical Significance
Vitamin D serum level in ng/mL	17.81± 8.11	26.61± 6.44	0.008	Sig.
CRP mg/dL	1.76±1.05	0.80± 0.00	0.000	Sig.
ESR mm/hr.	38.50±14.70	24.05± 10.38	0.010	Sig.

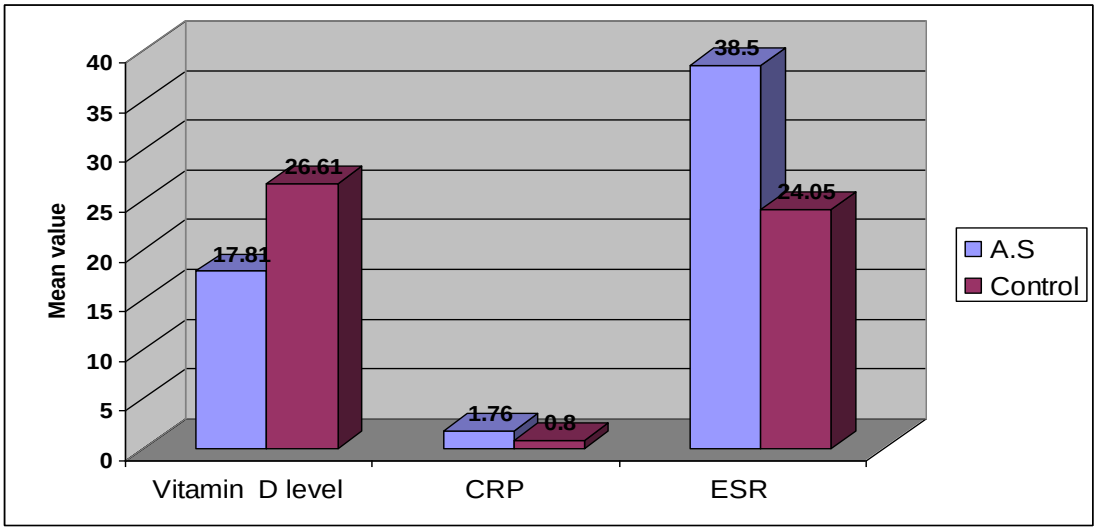

Fig. (14) Statistical comparison of vitamin D serum level, ESR and CRP between the target A.S patients and control group.

Table (13) Statistical comparison of vitamin D serum level ,ESR and CRP between the active A.S and the inactive A.S group:

Characteristic	**Active A.S group** ***(n = 10)*** **Mean ± S.D**	**Inactive A.S group** ***(n = 10)*** **Mean ± S.D**	**P-value**	**Statistical Significance**
Vitamin D serum level in ng/mL	14.83± 7.33	20.78± 8.48	0.175	N.S
CRP mg/dL	2.40± 1.13	1.12± 0.44	0.032	Sig.
ESR mm/hr.	49.80± 10.38	27.20± 7.69	0.012	Sig.

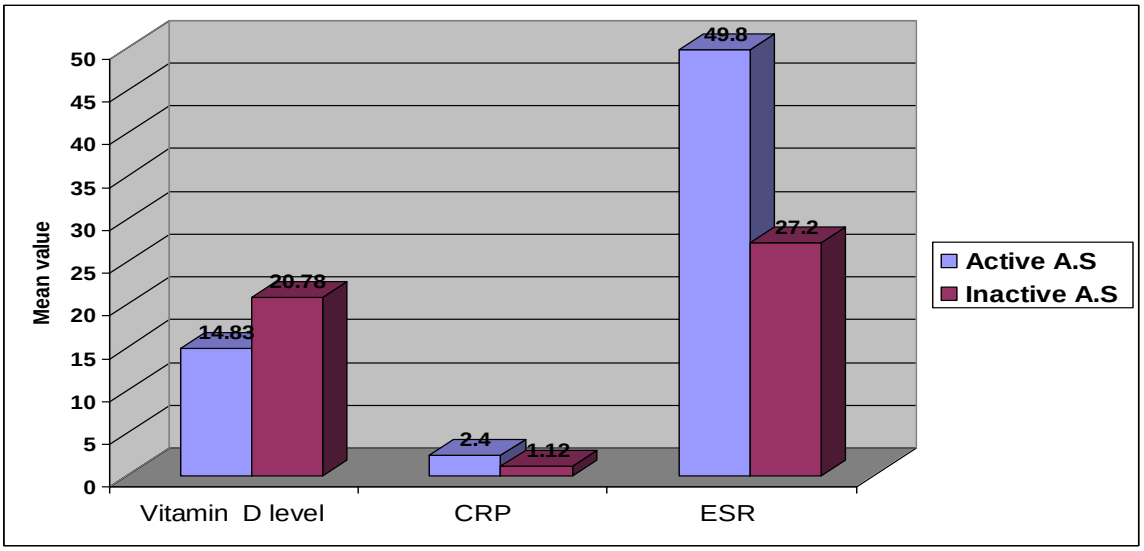

Fig. (15) Statistical comparison of vitamin D serum level, ESR and CRP between the active A.S and the inactive A.S group.

DISCUSSION

In the current study thirty patients with RA fulfilling ACR criteria for the classification of rheumatoid arthritis and twenty healthy controls were included.

The mean value of vitamin D serum level (ng/mL) was found to be low in R.A. group (Mean ± S.D)(13.47± 8.17) in comparison to the control group (26.61± 6.44). This difference was statistically significant. The comparison of vitamin D level between the active R.A. group and the inactive R.A. group did not show any statistically significant difference.

This finding was matched with GAIL et al., (2011) who reported that in elderly male RA patients 1. 25(OH) 2 D3 insufficiency was highly prevalent. On the other hand, there was a conflicting report by Rossini et al., (2010) who reported that vitamin D deficiency is quite common in RA patients, but similar to that found in control subjects; and disease activity and disability scores are inversely related to 1. 25(OH) 2 D3 levels.

This study was incompatible with Cutolo et al., (2009) who reported that no significant differences were found concerning 1. 25(OH) 2 D3serum level between RA patients and their controls in both North and South European RA patients, in addition, 1. 25(OH) 2 D3 values showed a significant negative correlation with RA clinical status (DAS28), suggesting possible effects of vitamin D among other factors on disease activity.

In the current study twenty SLE patients with mean age of 37.65 ± 12.37 years and twenty control cases were studied. The study shows vitamin D deficiency in SLE patients as the mean value of vitamin D level was (19.32 ±10.67) in the SLE group, while in the control group it was(26.61 ± 6.44). This difference in the mean value of vitamin D level between the target SLE group and control group was found to be statistically significant, as p-value of 0.019 and the difference in the mean value of vitamin D level between the SLE active group and inactive group was found to be statistically insignificant, as p-value of 0.470.

This finding was matched with the study done by Souto et al., (2011). Their objectives were to determine the prevalence of vitamin D insufficiency in Brazilian lupus patients and study the relationship between vitamin D insufficiency and disease activity ,the study included 159 SLE patients and showed that the prevalence of vitamin D insufficiency and deficiency were 37.7% and 8.2%, respectively, levels of 1. 25(OH) 2 D3 were not associated with lupus activity score which is compatible with our study. Similar study done by Bonakdar et al., (2011) showed that most of the SLE patients have vitamin D deficiency at the time of diagnosis that is associated with a higher disease activity. Another study was done by Borba et al., (2009); the association between vitamin D deficiency and disease activity was demonstrated and levels of 1. 25(OH) 2 D3 were lower (17.4 ± 12.5) in patients with high disease activity when compared to those with mild disease activity and the control group.

In a Spanish study was done by Ruiz-Irastorza et al., (2008) on 92 SLE patients, there were low levels of vitamin D (< 30 ng/mL) in 75% of the patients and deficiency (< 10 ng/mL) in 45% of them. 45% of the patients with low levels and 35% of those with deficiency were on calcium and vitamin D supplementation at the time of the evaluation.

In this current study, the relation between vitamin D serum level and the disease activity in patients with ankylosing spondylitis (AS) was investigated on ten patients with AS and twenty healthy individuals ~~were~~ included in the study. The study showed that: in the patient group, the vitamin D serum levels were lower than the control group ; (17.81 ± 8.11) and (26.61 ± 6.44) respectively. This was found to be statistically significant, as p-value of 0.008.

The difference in the mean value of vitamin D serum level between the A.S. active group and inactive group was found to be statistically insignificant as p-value of 0.17.

This finding was compatible with Bedriye et al., (2010) the vitamin D serum levels were lower in AS patients than in the control group.

In this current study ten patients with Behçet's Disease and twenty matched healthy controls were included. The diagnostic criteria for Behçet's Disease proposed by the American college of rheumatology were used for diagnosis.

In this study the mean value of vitamin D serum level in the Behçet's disease group was low in comparison to vitamin D serum level in the control group(17.64 ±8.79) and (26.61 ± 6.44) respectively.

The difference in the mean value of vitamin D serum level between the target Behcet disease group and control group was found to be statistically significant, as p-value of 0.041 .

In comparison of vitamin D serum level between the active Behçet's disease and the inactive Behcet disease group there was no statistically significant difference as the mean value of vitamin D serum level was (14.74 ±7.70) in the Behcet disease active group, while in the Behçet's disease inactive group it was(20.54 ± 9.66) and P-value 0.465.

This is compatible with Saliha et al., (2011) who reported that1. 25(OH) 2 D3 serum levels are decreased in patients with Behçet's Disease.

This study is also compatible with Christina et al., (2010) who reported that vitamin D deficiency occurs at a higher rate in patients with Behçet's Disease, thus appropriate supplementation should be indicated.

In the current study thirty O.A. patients with mean age of 62.27± 10.35 years and twenty control cases were studied. The difference in the mean value of vitamin D serum level between the target O.A. group and control group was found to be statistically insignificant, as p-value of 0.178 as the mean value of vitamin D serum level was(22.95 ±9.30) In the O.A. group, while in the control group it was (26.6 1 ± 6.44) .

The difference in the mean value of vitamin D serum level between the O.A. active group and inactive group was found to be statistically insignificant, as p-value of 0.430.

This finding was incompatible with the study done by Changhai Ding_et al., (2009) who reported that the serum level of 1. 25(OH) 2 D3 levels are associated with decreased knee cartilage loss (assessed by radiograph or MRI) in subjects with radiographic OA and knee pain.

Also it was not matched with Bergink et al., (2009) who reported that low vitamin D serum level and Low dietary vitamin D intake increases the risk of progression of knee OA. Thus, improving the vitamin D status in the elderly could protect against the development and worsening of knee OA, especially in those with low bone mineral density(BMD).

CONCLUSION

- Vitamin D is recognized as an important immunomodulatory factor involved in autoimmune rheumatic diseases.
- These immunomodulatory and anti-inflammatory activities might be particularly efficient in the treatment of rheumatic patients and support a therapeutic role of 1, 25(OH)2D3 in such diseases.
- Vitamin D deficiency occurs at a higher rate in patients with autoimmune disorders such as RA ,SLE , Behcet disease and A.S.
- Routine screening for vitamin D deficiency in early rheumatic diseases is recommended.
- A much higher oral vitamin D intake than recommended in current guidelines is safe and necessary to maintain adequate circulating 1, 25 (OH)2D3 levels especially in the absence of UVB radiation to the skin.
- Further studies should be performed on a larger number of rheumatic patients to a the role vitamin D in rheumatic diseases and its relation to disease activity.

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