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Pitout JDD, Church DL, Gregson DB, Chow BL, McCracken M, Mulvey M, Laupland KB (2007).

Molecular epidemiology of CTXM-producing Escherichia coli in the Calgary Health Region: emergence of CTX-M-15-producing isolates.

Antimicrob. Agents Chemother. 51: 1281-1286.

Pelczar JR, Harley JP, Klein DA (1993). Microbiology: Concepts and Applications. McGraw-Hill Inc., New York, pp. 591-603.

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# International Journal of Nutrition and Metabolism

Full Length Research Paper

# Relationship between vitamin D and disease activity in some rheumatic diseases

Mohamed Hashem Ibrahim<sup>1</sup>, Mohamad Sayyed Bakheet<sup>2</sup>, Khaled A Abdel-Sater<sup>3</sup> and Maher Abdel shakoor<sup>3</sup>

<sup>1</sup>Rheumatology, Physical Medicine and Rehabilitation Department, Makka, Kingdom of Saudi Arabia.

<sup>2</sup>Biochemistry Department, Al-Azhar Faculty of Medicine, Assiut, Egypt.

<sup>3</sup>Physiology Department, Al-Azhar Faculty of Medicine, Assiut, Egypt.

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Most people are aware that vitamin D deficiency in young children can lead to rickets, a condition where the bones become weak and soft. Many of the benefits of vitamin D relate to its role in the modulation of immune system. So, vitamin D may play role in autoimmune rheumatic diseases. There are two groups: patients group and control group. The control group included 20 healthy volunteers. Patients group included 100 rheumatic patients, 30 with rheumatoid arthritis (RA), 20 with systemic lupus erythematosus (SLE), 30 with osteoartheritis (OA), 10 with Behcet's disease, 10 with ankylosing spondylitis (AS). Venous blood samples were taken for determination of erythrocyte sedimentation rate (ESR), serum 1. 25(OH) 2 D3 levels and serum C reactive protein (CRP) levels. The disease activity in different target groups was assessed using Disease Activity Score including 28 joint counts (DAS28) in RA patients, SLE disease activity index (SLEDAI) in SLE patients, Western Ontario and McMaster Universities Arthritis Criteria (WOMAC) in OA patients, The American College of Rheumatology (ACR) criteria in Behcet's patients and Bath Ankylosing Spondylitis Disease Activity Index (BASDAI) in A.S. patients. The mean value of vitamin D serum levels were significantly lower in each of RA, SLE, Behcet's disease and AS patients (mean ± SD) (13.47 ± 8.17, 19.32 ±10.67, 17.64 ±8.79 and 17.81 ±8.11 respectively) in comparison to control group (26.61 ± 6.44, p-value ≤0.05). While, There is no significance difference between the OA (22.95 ±9.3) and control groups, as p-value of 0.178. As regard the comparison of vitamin D serum level between the active RA, SLE, OA, Behcet's disease and AS patients and the inactive groups, the difference was found to be statistically insignificant (p-value >0.05). In the patients group DAS28 in RA patients, SLEDAI in SLE patients, WOMAC in OA patients, ACR criteria in Behcet's patients and BASDAI in AS were significantly higher in active groups as compared with inactive groups. In the present study no association was observed between vitamin D levels and disease activity scales DAS28, SLEDAI, WOMAC, and ACR, While there is a significant negative correlation between vitamin D levels and BASDAI. Vitamin D deficiency occurs at a higher rate in patients with RA, SLE, Behcet's disease and A.S. While, no association was observed between vitamin D levels and disease activity scales in the RA, SLE, OA, and Behcet's disease patients.

Key words: Vitamin D, immune system, rheumatic autoimmune diseases, activity and severity.

#### INTRODUCTION

Vitamin D is one of the fat soluble vitamins derived from Cholecalciferol (7-dehydrocholesterol, pro-vitamin  $D_3$ ) in

humans and from ergosterol (pro-vitamin  $D_2$ ) in yeast and plants, both forms are bioactive. The main source of

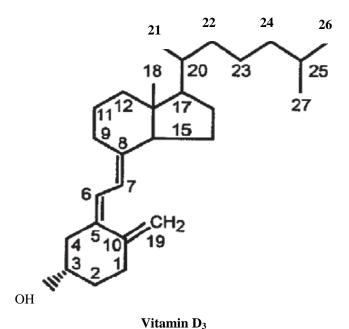


Figure 1. Cholecalciferol (D3) (Adams, and Hewison, 2010).

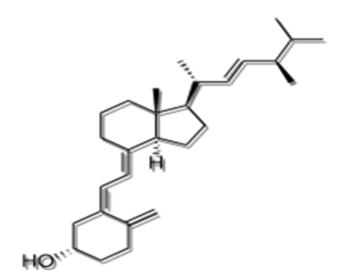


Figure 2. Ergocalciferol (D2) (Joshi et al., 2010).

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) Figure 1 and 2. Vitamin  $D_3$  (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  $\alpha$ -globulin (Cheng et al., 2004).

Calcidiol is then converted in the kidneys (by the enzyme  $25(OH)D-1\alpha$ -hydroxylase) into calcitriol (1,25- $(OH)_2D_3$ ), 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) Figure 3.

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) Figure 4.

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  $D_3$ , the biologically active metabolite of Vitamin  $D_3$ , 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) Figure 5, Table 1.

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

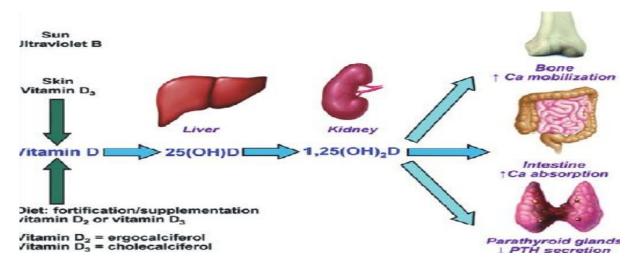
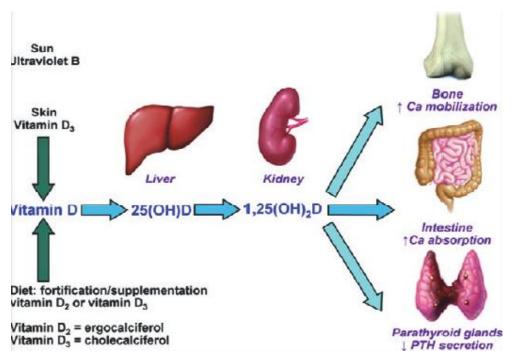


Figure 3. Mechanism of synthesis of vitamin D: 1, 25 dihydroxyvitamin D (Cheng et al., 2004).



**Figure 4.** Role of synthesis and metabolism of Vitamin D in the regulation of calcium and bone mineralization (Holick et al., 2007).

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.

Low serum levels of vitamin  $D_3$  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 (that is, 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,

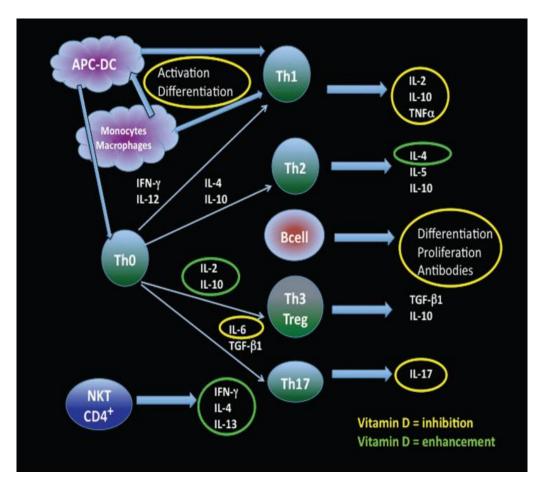


Figure 5. Mechanisms involved in vitamin D modulation of the immune responses. Dendritic cells (DCs) are primary targets for the immunomodulatory activity of 1,25(OH)2D3, as indicated by inhibited DC differentiation and maturation, together with inhibition of differentiation of monocyte precursors into 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 CD4\(\beta\)-helper cell subsets (Th1, Th2, Th3-Treg, Th17) originating from nai 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).

and none have assessed the association of vitamin D with disease onset. Merlino et al. (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  $\alpha$  (TNF $\alpha$ ) 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 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 D3 in patients with undifferentiated connective tissue disease (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,  $14.7 \pm 6.45$  ng/ml versus  $33.0 \pm 13.4$  ng/ml, P = 0.0001, respectively.

**Table 1.** Demographic data and duration of illness in the patients group.

Parameter	RA group (n=30) Mean±SD	SLE group (n=20) Mean±SD	OA group (n=30) Mean±SD	Behcet disease group (n=10) Mean±SD	AS group (n = 10) Mean±SD	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	<0.001	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	< 0.001	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 2. Disorders that have been linked to 1. 25(OH) 2 D3.

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

(Cutolo and Otsa 2008).

Prospective studies available for the 4 major autoimmune diseases: RA, SLE, MS, and type 1 diabetes mellitus (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  $1,25(OH)_2D_3$  in different rheumatic diseases to find the relation between  $1,25(OH)_2D_3$  level and rheumatic diseases and to establish its relation to the rheumatic diseases activity and severity.

#### MATERIALS AND METHODS

This study was done on 100 patient selected from the outpatient clinic of rheumatology department faculty of medicine, Alazher university, Assuit branch as patients group, their age ranged between 16 to 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 matched the patients group in age and socio-economic status. The study has been approved by the relevant research and ethics committee after informed consent for each of patients and control groups.

Patients group was subdivided into: 30 patients suffering from RA, 30 patients suffering from OA, 20 patients suffering from SLE, 10 patients suffering from Behcets disease and 10 patients suffering from ankylosing spondylitis. The following iwere done for each patients and control groups:

- 1. 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.
- 2. Venous blood samples were taken for determination of complete blood count using automated cell counter, ESR using westergren tubes method, serum  $1,25(OH)_2D_3$  level, serum calcium (total and ionized), serum phosphorus, serum parathormone, blood urea and serum

**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	$40.30 \pm 7.83$	41.90 ± 11.70	0.000	Sig
Age of onset (years)	32.57 ± 8.23	$27.35 \pm 8.80$	49.30 ± 15.67	$26.70 \pm 5.03$	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	$8.70 \pm 4.69$	0.975	N.S.

NS = non significant.

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 $\pm$ 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	$0.80 \pm 0.00$	0.000	Sig.
ESR mm/hr.	53.00± 24.17	24.05± 10.38	0.000	Sig.

creatinine, liver function tests (aspartate aminotransferase and alanine aminotransferase, AST and ALT), rheumatoid factor using latex agglutination test, anti-nuclear antibodies (ANA) and anti double stranded DNA and anti Sm ab, fasting and post prandial blood glucose level and complete urine analysis by microscopic examination.

- 3. Radiology: Plain X-ray of hands and feet (postero-anterior view).
- 4. Plain X-ray of other affected joints.
- 5. The disease activity in different target groups was assessed using disease activity score 28 (DAS28) in RA patients, SLE disease activity index (SLEDAI) in SLE patients, bath ankylosing spondylitis disease activity index (BASDAI) in AS patients, The Western Ontario and McMaster Universities Arthritis Index (WOMAC osteoarthritis index) in OA patients and American College of Rheumatology (ACR) criteria in Behcet patients.

Assessment of  $1,25(OH)_2D_3$  sufficiency by *in vitro* quantitative determination of  $1,25(OH)_2D_3$  in human serum was done 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 which 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 2010). ECLIA is based on competition principle. Total duration of assay is 18 min.

**1st incubation:** 1,25(OH)<sub>2</sub>D<sub>3</sub> in the sample (35  $\mu$ I) competes with the biotin labeled vitamin D in the complex contained in R2 (biotin-vitamin D/polyclonal 1,25(OH)<sub>2</sub>D<sub>3</sub> - specific ruthenium labeled antibody). The remaining amount of the complex (biotin-vitamin D/polyclonal 1,25(OH)<sub>2</sub>D<sub>3</sub> - specific ruthenium labeled antibody) is dependent upon the analyte concentration in the sample.

incubation: After streptavidin-coated 2nd addition of 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 2point 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:  $nmol/l \times 0.40 = ng/ml$ 

or  $ng/ml \times 2.50 = nmol/L$ 

Data were expressed as mean  $\pm$  standard deviation. Comparisons were performed for normal distributed data using t-test for independent groups. P-value considered insignificant if > 0.05, significant if < 0.05, highly significant if < 0.001, the statistical analysis were done using statistical package for social sciences (SPSS) V11.0 program Table 3 to 13, Figure 6 to 15.

#### **RESULTS AND 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 RA group (mean  $\pm$  SD) (13.47  $\pm$  8.17) in comparison to the control group (26.61  $\pm$  6.44). This difference was statistically significant. The comparison of vitamin D level

**Table 5.** Statistical comparison of serum vitamin D, CRP and ESR levels between the target SLE patients and control groups.

Characteristic	SLE group (n=20) Mean±SD	Control group (n = 20) Mean±SD	P-value	Statistical Significance
Vitamin D ng/mL	19.32± 10.67	26.61± 6.44	0.019	Sig.
CRP mg/dL	3.60±3.04	$0.80 \pm 0.2$	< 0.001	Sig.
ESR mm/hr.	51.30± 19.15	24.05± 10.38	< 0.001	Sig.

**Table 6.** Statistical comparison of serum vitamin D, CRP, ESR and SLEDAI levels between the active SLE group and the inactive SLE group.

Characteristic	Active SLE group (n=5) Mean±SD	Inactive SLE group (n=5) Mean±SD	P-value	Statistical significance
Vitamin D ng/ml	18.33± 3.1	21.87± 2.07	0.346	N.S.
CRP mg/dl	$5.88 \pm 0.69$	1.16± 0.28	< 0.001	Sig
ESR mm/hr.	64.00± 5.27	38.47± 4.3	0.003	Sig.
SLEDAI	4.98±0.62	1.62±0.27	< 0.001	Sig.

**Table 7.** The relation between vitamin D values and disease activity parameters in the SLE group.

Characteristic	r	P-value	Statistical Significance
CRP mg/dL	0.35	0.35	N.S
ESR mm/hr.	- 0.12	0.75	N.S
SLEDAI	- 0.22	0.56	N.S

Table 8. Statistical comparison of serum vitamin D, CRP and ESR between the target OA patients and control group.

Characteristic	OA group (n = 30) Mean ± S.D	Control group (n = 20) Mean ± S.D	P-value	Statistical significance
Vitamin D ng/mL	22.95± 9.30	26.61± 6.44	0.178	N.S.
CRP mg/dL	1.25±0.80	$0.80 \pm 0.00$	< 0.001	Sig.
ESR mm/hr.	35.00±13.72	24.05± 10.38	0.006	Sig.

**Table 9.** Statistical comparison of serum vitamin D, CRP, ESR and WOMAC levels between the active OA and the inactive OA group.

Characteristic	Active Behcet disease group (n=5) Mean±SD	Inactive Behcet disease group (n=5) Mean±SD	P-value	Statistical Significance
Vitamin D ng/ml	21.874±2.07	24.89±2.65	0.38	NS
CRP mg/dl	1.60±0.70	0.95±0.32	0.003	Sig.
ESR mm/h	43.93±10.45	27.19±11.39	<0.001	Sig.
WOMAC	1.31±0.17	0.72±0.11	0.013	Sig.

between the active RA group and the inactive RA 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  $1,25(OH)_2D_3$  insufficiency

Table 10. The relation between vitamin D values and disease activity parameters in the OA group.

Characteristic	r	P-value	Statistical Significance
CRP mg/dL	0.35	0.35	Sig.
ESR mm/hr.	- 0.12	0.75	Sig.
WOMAC	- 0.22	0.56	Sig.

**Table 11.** Statistical comparison of vitamin D serum level, ESR and CRP between the target Behcet's disease patients and control group

Characteristic	Behcet's disease group (n = 10) Mean ± S.D	Control group (n = 20) Mean ± S.D	P-value	Statistical Significance
Vitamin D ng/mL	17.64± 8.79	26.61± 6.44	0.006	Sig.
CRP mg/dL	1.85±1.76	$0.80 \pm 0.2$	< 0.001	Sig.
ESR mm/hr.	38.70±18.51	24.05± 10.38	0.041	Sig.

**Table 12.** Statistical comparison of serum vitamin D, CRP, ESR and ACR levels between the active Behcet's disease and the inactive Behcet's disease group:

Characteristic	Active Behcet's disease group (n = 5) Mean ± S.D	Inactive Behcet's disease group (n = 5) Mean ± S.D	P-value	Statistical Significance
Vitamin D 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.
ACR	$3.72 \pm 0.60$	$1.53 \pm 0.20$	< 0.001	Sig.

**Table 13.** The relation between vitamin D values and disease activity parameters in the Behcet's disease group:

Characteristic	r	P-value	Statistical Significance
CRP mg/dL	0.07369	0.85	N.S
ESR mm/hr.	-0.05468	0.89	N.S
ACR	-0.2797	0.47	N.S

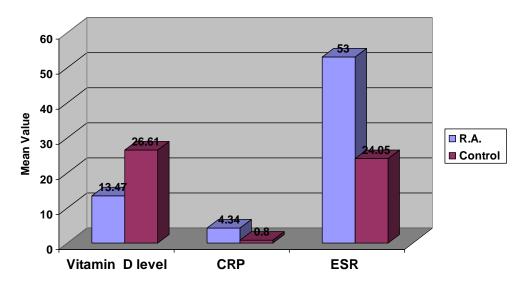
**Table 14.** Statistical comparison of vitamin D serum level, ESR and CRP between the target AS patients and control group

Characteristic	A.S group (n = 10) Mean ± S.D	Control group (n = 20) Mean ± S.D	P-value	Statistical Significance
Vitamin D ng/mL	17.81± 8.11	26.61± 6.44	0.008	Sig.
CRP mg/dL	1.76±1.05	$0.80 \pm 0.00$	< 0.001	Sig.
ESR mm/hr.	38.50±14.70	24.05± 10.38	0.010	Sig.

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 ng/mL	14.1± 1.07	18± 2.13	0.121	N.S
CRP mg/dL	2.43± 0.14	1.32± 0.15	<0.001	Sig.
ESR mm/hr.	47.8± 3.65	26.48± 2.59	<0.001	Sig.
BASDAI	5.61 ± 0.76	$3.28 \pm 0.56$	0.018	Sig.

Table 15. The relation between vitamin D values and disease activity parameters in the AS group.

Characteristic	r	P-value	Statistical Significance
CRP mg/dL	-0.17	0.66	N.S
ESR mm/hr.	- 0.07	0.84	N.S
BASDAI	- 0.72	0.03	Sig.



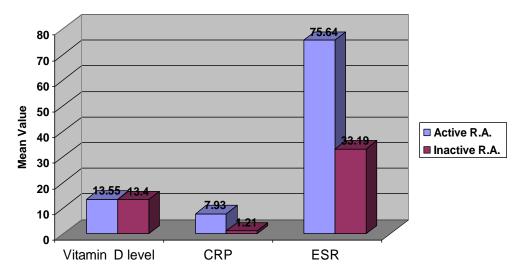
**Figure 6.** Statistical comparison of vitamin D serum level, ESR and CRP between the target R.A. patients and control group.

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 concerning  $1,25(OH)_2D_3$  serum level between RA patients and their controls in both North and South European RA patients, in addition,  $1,25(OH)_2D_3$  values showed a significant negative correlation with RA clinical status (DAS28), suggesting possible effects of vitamin D among other factors on disease activity.

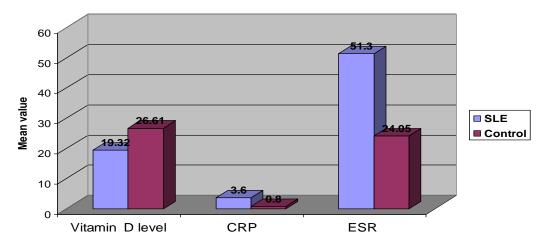
In the current study, twenty SLE patients with mean age of  $37.65 \pm 12.37$  years and twenty control cases were studied. The study shows vitamin D deficiency in SLE patients as the mean value of vitamin D level was  $(19.32 \pm 10.67)$  in the SLE group, while in the control group it was  $(26.61 \pm 6.44)$ . 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.

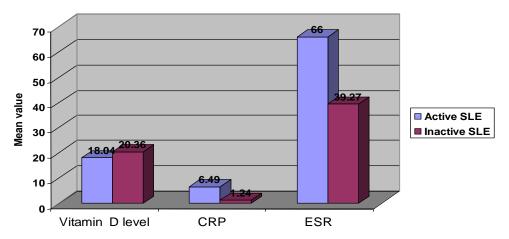
This finding was matched with the study done by Souto et al. (2011). Their objectives were to determine the prevalence of vitamin D insufficiency in Brazilian lupus patients and study the relationship between vitamin D insufficiency and disease activity, the study included 159 SLE patients and showed that the prevalence of vitamin D insufficiency and deficiency were 37.7 and 8.2%, respectively, levels of 1,25(OH)<sub>2</sub>D<sub>3</sub> were not associated with lupus activity score which is compatible with our study. Similar study done by Bonakdar et al. (2011)



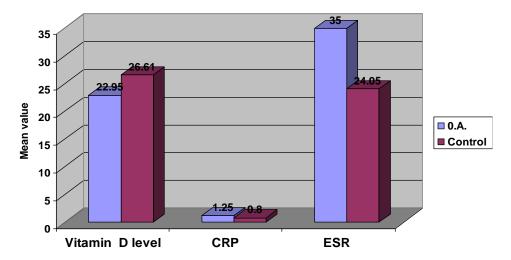
**Figure 7.** Statistical comparison of vitamin D serum level, ESR and CRP between the active R.A. group and the inactive RA group.



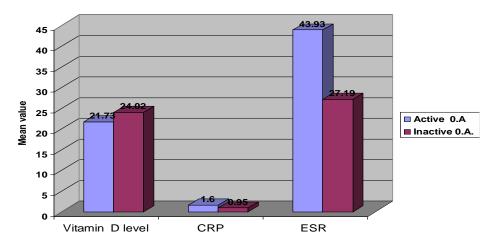
**Figure 8.** Statistical comparison of vitamin D serum level, ESR and CRP between the target SLE patients and control group.



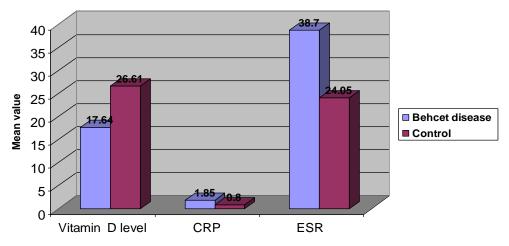
**Figure 9.** Statistical comparison of vitamin D serum level, ESR and CRP between the active SLE group and the inactive SLE group.



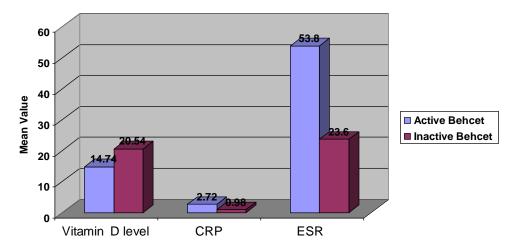
**Figure 10.** Statistical comparison of vitamin D serum level, ESR and CRP between the target 0.A. patients and control group.



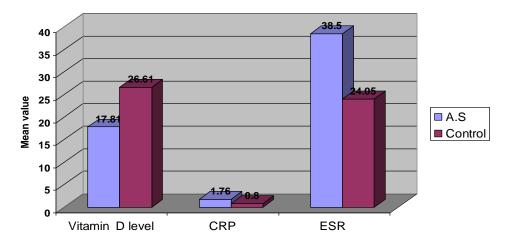
**Figure 11.** Statistical comparison of vitamin D serum level, ESR and CRP between the active OA and the inactive OA group.



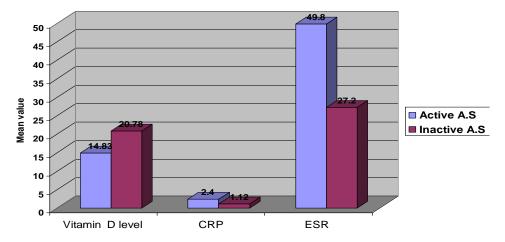
**Figure 12.** Statistical comparison of vitamin D serum level, ESR and CRP between the target Behcet disease patients and control group.



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



**Figure 14.** Statistical comparison of vitamin D serum level, ESR and CRP between the target AS patients and control group.



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

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  $1,25(OH)_2D_3$  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 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 in ten patients with AS and twenty healthy individuals 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  $\pm$  8.11) and (26.61  $\pm$  6.44), respectively. This was found to be statistically significant, with p-value of 0.008. The difference in the mean value of vitamin D serum level between the AS 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) who found out that the vitamin D serum levels were lower in AS patients than in the control group.

In this current study, ten patients with Behcet'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  $(17.64 \pm 8.79)$  and  $(26.61 \pm 6.44)$ , 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, with pvalue of 0.041. In comparison with vitamin D serum level between the active Behcet's disease and the inactive Behcet disease group, there was no statistically significant difference as the mean value of vitamin D serum level was  $(14.74 \pm 7.70)$  in the Behcet disease active group, while in the Behçet's disease inactive group it was  $(20.54 \pm 9.66)$  and p-value 0.465. This is compatible with Saliha et al. (2011) who reported that 1,25(OH)<sub>2</sub>D<sub>3</sub> 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 OA patients with mean age of  $62.27 \pm 10.35$  years and twenty control cases were studied. The difference in the mean value of vitamin D serum level between the target OA group and control group was

found to be statistically insignificant, with p-value of 0.178 as the mean value of vitamin D serum level was (22.95  $\pm$  9.30) in the OA group, while in the control group it was (26.6 1  $\pm$  6.44). The difference in the mean value of vitamin D serum level between the OA active group and inactive group was found to be statistically insignificant, with p-value of 0.430.

This finding was incompatible with the study done by Changhai et al. (2009) who reported that the serum level of 1,25(OH)<sub>2</sub>D<sub>3</sub> 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)<sub>2</sub>D<sub>3</sub> in such diseases. Vitamin D deficiency occurs at a higher rate in patients with autoimmune disorders such as RA, SLE, Behcet disease and AS. 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)<sub>2</sub>D<sub>3</sub> levels especially in the absence of UVB radiation to the skin. Further studies should be performed on a larger number of rheumatic patients to the role of vitamin D in rheumatic diseases and its relation to disease activity.

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# International Journal of Nutrition and Metabolism

Full Length Research Paper

# Study of some chemical, physical, sensory and bacteriology characteristics of canned chicken meat imported to Sulaymaniyah markets, Iraq

#### **Hemin Nuradden Mohammed**

Department of Animal production/Faculty of Agricultural Sciences/Sulaimani University, Iraq.

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This study was conducted to assess the quality of imported four brands of canned chicken meat that were (A, B, C and D) by using different quality standard inspection tests; these are, determination the chemical compositions of moisture, protein, fat, ash, energy and as well as studying the changes in the organoleptic characters represented by measuring peroxide value, free fatty acids, thiobarbituric acid and total volatile nitrogen were carried out. The microbiological investigations involved examination of total bacteriological count, coliform bacteria, proteolytic, lipolytic and sporoforming bacteria (anaerobic) also were tested. In addition, sensory attributes were measured. There are significant differences (p ≤ 0.05) among trademarks in chemical analyses that indicated that A trademark of canned chicken meat had the highest percentage of moisture (67.05%) compared to the lowest percentages of D trademark (59.31%) and high protein contents were in B (32.10%) trademarks, while lower contents were in A 20.72% trademark. B trademarks contained low percentage of lipid (2.69%), while D trademark contained a high percentage of lipids (12.53%). C trademark appeared to have higher contents of ash (3.40%), while lower contents were in A (2.66%) trademark, and the total volatile nitrogen values for the all trademarks were non-significant. There were no significant differences (p  $\leq$  0.05) among all trademarks in contents of free fatty acids and thiobarbituric acid (TBA), but there are significant differences (p ≤ 0.05) among all trademarks in contents of peroxide values. Peroxide values (PV) for A, B, C and D trademarks were 0.95, 0.45, 0.65 and 0.80 meg oxygen/kg fat, respectively. Also, PV was through the allowance limits for all trademarks. There are not significant differences (p ≤ 0.05) in microbial tests among all trademarks that indicated there were no aerobic bacteria in any of these trademarks. Significant differences in the sensory properties among the four trademarks observed, while there are non-significant differences in the overall acceptability of the four trademarks.

**Key words:** Canned chicken meat, quality analysis, sensory evaluation.

#### INTRODUCTION

Food composition data are important to a spectrum of users ranging from international organizations and private individuals, to food assistance programs, epidemiologists correlate patterns of disease with dietary components

and nutritional assessment of individual intake and dietetic counseling (Rand et al., 1991). Applying food safety standards on a product is very important because it relates closely to human's health. Good food products

E-mail: hemn33@yahoo.com.

have a high nutritional quality, as well as being free from physical, chemical and biological contaminations. The food industry development encourages food manufacturer's to produce more practical and durable products, but still must have high nutrition. For example, beef processing to produce meatballs, corned beef, beef burgers and sausages have the purpose to form more practical and durable products, as well as having high nutritional value (Farmer and Farmer, 2000; Javed et al., 2009).

Chicken meat can make many positive contributions to the diet of those on low incomes. Although not all meat is seen as healthy, chicken meat is, and is frequently more affordable than other meats. It is of a consistently high quality, is low in saturated fats, can be enriched with some essential nutrients and is sought after worldwide (Yu et al., 2008; Bingham, 2006). Chicken meat does not contain the trans fats that contribute to coronary heart disease, and can be found in high amounts in beef and lamb. In Canada, values of 2 to 5 percent have been reported for beef and as high as 8 percent for lamb. The World Cancer Research Fund and others (Bingham, 2006) and Acuff (2006) clarified the difference between spoilage organisms and pathogens by stating, "spoilage organisms will not make you sick, as in instigating an infection and creating a real illness." However, spoilage organisms make food undesirable. The meat industry works diligently to prevent, reduce and eliminate both pathogenic and spoilage bacteria before meat are delivered to consumers for purchase.

Canned luncheon meat is an emulsion-type, cured meat product that is sterilized by heat and has a shelf-life of about three years at ambient temperature (Standard, 1998). It is a popular food item in many countries and it is also used in 'fast food' (Al-Bachir and Mehio, 2001). The basic raw material is either beef or poultry in chopped or comminuted form, and additional ingredients may include spices, soya protein, starch, nitrite, salt, ascorbate, and phosphate (Abdullah, 2007). Meat can be contaminated with foodborne human pathogens and is a highly perishable type of food; heat treatment of the canned product is essential in relation to its safety and stability, and must be sufficient to ensure that no microbiological hazard arises during storage (Ostoja et al., 2002). The quality of luncheon products is strongly influenced by the temperature, time of processing and fat content of the meat. If too severe, heat treatment can cause denaturation of protein and changes in product appearance, water-binding capacity and tenderness (Pena-ramos and Xiong, 2002).

As one of the ways to keep safety of food in Sulaymaniyah-Iraq, this study aims to assess the quality of canned meat by parameters used in quality control included sensory, physical, chemical, microbiological, also having knowledge about International and national food laws of meat and poultry act, prevention of food adulteration Act and food additives.

#### **MATERIALS AND METHODS**

#### Sampling

This study was conducted in the laboratories of Faculty of Agricultural Science and Quality Control Laboratories of Veterinary Directorate. Samples included canned chicken meat to four different brands, A, B, C and D. The brands are most commercially available in Sulaymaniyah governorate. The total number of samples used in the study was 24 samples of 6 replicates for each brand, and taken into account when the acquisition of samples was done to date.

#### Moisture content

Moisture content was observed according to the method of Association of Official Analytical Chemistry (AOAC, 2000).

#### Ash content

Ash percentage was determined by Gravimetric method as described by AOAC (2000).

#### **Total protein content**

Protein content was determined according to the method as described by AOAC (2000).

#### Fat content

Total fat content was extracted in Soxhlet extraction unit as described by AOAC (2000).

#### Calculation of caloric value

The caloric value of 100 g meat was calculated according to Atwater and Woods (1986).

#### Free fatty acids (FFA)

This was estimated by the way of Egan et al. (1981).

#### Thiobarbituric acid (TBA)

The value analysis was measured by the way of Tarladgis et al. (1960) as adopted by Witte et al. (1970).

#### Peroxide value (PV)

This was analyzed by the way of Egan et al. (1981).

#### Total volatile nitrogen (TVN)

This was estimated by the way of Malle and Poumeyrol (1989).

#### **Bacteriological analyses**

#### Sample preparation

For the microbiological analysis of all trademarks, 25 g of samples

Table 1. F	valuation form	for descriptions	s of sensory p	roperties for all tr	ademarks

Overall acceptability	Color	Flavor	Juiciness	Tenderness
(5) Very acceptable	(5) Very dark	(5) Very good	(5) Very juice	(5) Very soft
(4) Acceptable	(4) Dark	(4) Good	(4) Juice	(4) Soft
(3) Middle	(3) Acceptable	(3) Middle	(3) Middle	(3) Middle
(2) Unacceptable	(2) Light	(2) Weak	(2) Dry	(2) Hard
(1) Rejected	(1) Very light	(1) Very weak	(1) Very dry	(1) Very hard

Table 2. Chemical analysis of four canned chicken meat imported to sulaymaniyah markets.

Trademarks	Moisture%	Dry matter%	Protein%	Fat%	Ash%	Energy
Α	67.05±0.98 <sup>a</sup>	32.94±0.9 <sup>b</sup>	20.72±2.36°	6.97±3.34 <sup>ab</sup>	2.66±0.0 <sup>c</sup>	170±1.96 <sup>b</sup>
В	59.93±2.53 <sup>b</sup>	40.07±2.5 <sup>a</sup>	32.10±0.18 <sup>a</sup>	2.69±2.40 <sup>b</sup>	3.22±0.0 <sup>b</sup>	160±2.25 <sup>c</sup>
С	60.18±0.07 <sup>b</sup>	39.81±0.0 <sup>a</sup>	28.18±0.42 <sup>ab</sup>	6.22±0.42 <sup>ab</sup>	$3.40\pm0.0^{a}$	176±2.17 <sup>b</sup>
D	59.31±0.42 <sup>b</sup>	40.68±0.42 <sup>a</sup>	23.59±0.62 <sup>bc</sup>	12.53±1.10 <sup>a</sup>	3.22±0.0 <sup>b</sup>	212±2.33 <sup>a</sup>

Means having the same letter in the same sections are not significantly different at  $P \le 0.05$ .

taken from different parts of the canned meat, was homogenized using a Waring blender at 6000 rpm in 225 ml of sterile salt solution (0.85% NaCl). All the tests performed on the samples were determined by the power plate technique. Decimal dilutions were prepared, and then by using a pipette 1 ml of each dilution was put into separate, duplicate, sterilized and appropriately marked petridishes, the petridishes were incubated but in a reverse manner. Finally, the colonies were calculated. The whole procedure was done according to (APHA, 1984). The performed tests are as follows:

**Total viable aerobic count:** The aerobic bacteria were enumerated on nutrient agar (Himedia labs. Pvt. Ltd) incubated at 35°C for 48 h.

**Total coliform bacterial counts:** Coliforms were determined on MacConkey agar containing bile salts (Himedia labs Pvt. Ltd) incubated at 37°C for 48 h.

**Proteolytic bacterial counts:** Proteolytic bacteria were determined using nutrient agar medium plus 10% sterilized skim milk. The plates were inoculated with the diluted sample homogenated and incubated at 30°C for 72 h and examined for clear zone around growth to indicate proteolytic activity.

**Lipolytic bacterial counts:** They were determined using nutrient agar medium plus 10% sterilized olive oil and plates were incubated at 30°C for 48 h. The lipolytic colonies were identified by copper sulphate 20% where blue colonies were counted.

**Total sporoforming bacterial counts:** Enumeration is carried out for bacteria belonging to species of (*Clostridium* and *Bacillus*), where the former is anaerobic while the latter is aerobic, using diluted solution 10<sup>-1</sup> and 10<sup>-2</sup> and were heated to 80°C for 10 min. Then, 1 ml of each diluted solution was transferred to a sterilized petridish. Consequently, sterilized nutrient agar was added and incubation was done as suitable for each bacterium. The plates were incubated for *Clostridium* in anerobic circumstances and at 37°C for 72 h while for *Bacillus* species, they were incubated aero-

bically at 35°C for 48 h.

#### Sensory evaluation

Sensory evaluation was carried out by a nine-member semi trained panel. Panel members with ages ranging from 25 to 50 were from faculty members and graduate students of Animal production Department of Sulaimani University, Faculty of Agricultural Science and all were experienced in sensory evaluation of various food products. Panelists were asked to evaluate the samples of each brand for tenderness, juiciness, flavor, color and overall acceptability as described in Table 1. The descriptions of sensory properties and how to rate a sample for the particular sensory property were on the evaluation form.

#### Statistical analysis

All data were subjected to one-way analysis of variance (ANOVA) using XL Stat program for windows. The level of significance was chosen at  $P \le 0.05$  and the results are presented as mean  $\pm$  SE. Duncan's multiple range tests was used to determine the significance of differences among means (Duncan, 1955).

#### **RESULTS AND DISCUSSION**

Moisture, dry matter, protein, fat, ash and energy contents of canned chicken meat are described in Table 2. There were significant differences (p < 0.05) in the chemical composition (moisture, dry matter, protein, fat, ash and energy) amongst the four trademarks of canned chicken meat examined. A contained high percentage of moisture (67.05%), while D contained low percentage (59.31%); however there are not significant difference between B, C and D. It is clear from the same table that the percentage of dry matter was on the exact opposite proportion of moisture, the highest ratios had been achieved in canned chicken meat containing the lowest

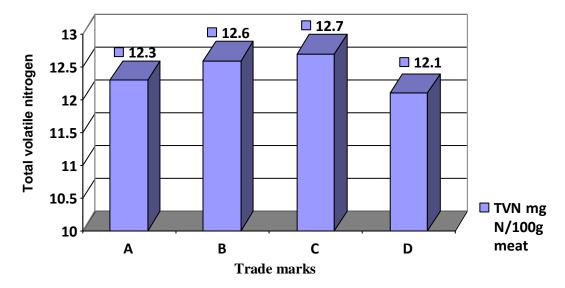


Figure 1. Total volatile nitrogen (TVN) value for four trademarks (mg N/100 g).

percentage of dry matter so that the moisture content and dry matter will be as a whole constituent of 100%. The percentage of moisture in the canned meat of A was high on the permissible limits by the Central Agency for Standardization and Quality Control (1988) but B, C and D trademarks was within the allowed limits. There were significant differences (p < 0.05) in the protein contents that the High protein contents were shown in B and C trademarks while lower contents were in A trademark.

A and D trademarks contained a moderate percentage (20.75 and 23.59%, respectively) of protein. Some studies reported similar protein content in canned meat, Alobaidi (2005) recorded the range of protein between 20.28 and 21.17% in canned meat. Romans and Ziegler (1977) found that the percentage of protein in fresh meat was 20% and in the canned meat, 22%. Thomas and Corden (1977) stated the chemical composition of different types of food, noticed that the percentage of protein in the canned meat was 20.9%. The proportion of protein in the majority of transactions are comparable to the minimum allowed, which amounts to 21% and this is not acceptable that we need a relatively high-protein sources to increase the protein consumption locally.

There were significant differences (p < 0.05) in the fat contents that the B trademarks contained low percentage of fat (2.69%) while D trademark contained high percentage of fat (12.53%). Fat in the canned meat samples were within the limits allowable of the Central Agency for Standardization and Quality Control (1988). The Central Agency for Standardization and Quality Control (1988) recorded that the percentage of fat in luncheon meat must not be greater than 25%. The results that are obtained here are within the range of fat content determined by many researchers as being 0.37 to 8% (Abeni and Bergoglio, 2001; Al-Najdawi and Abdullah, 2002; Van Heerden et al., 2002; Wattanachant et al.,

2004; Chuaynukool et al., 2007). Nevertheless, the differences in fat content in the inspected samples could be due to the differences in genetic and non-genetic factors (Lin et al., 1980; Bogosavljevic-Boskovic et al., 2010).

In conclusion, fat content of all samples were within the fat ranges that have been published by many researchers, but from the nutrition site, the fat content in all inspected samples were higher than what is being specified by United States Department of Agriculture (USDA) (2010). Significant difference in ash was shown among the four trademarks of canned meat. C appeared that have higher contents of ash that was higher than the permissible limits by Alobaidi (2005) that recorded the range of ash between 2.55 and 2.95%. Ash content was high in some samples, especially in the sample C that content 3.40%; as the ash content is an indication of the content of salts, it might indicate that preservatives present are salts in concentrations higher than specified. The ratio of carbohydrates came within the limits allowable which must not exceed 2%. The differences of ash content among the trade marks for all samples may be due to the decrease of moisture content which is associated with storage and handling proceedings with extension in storage period (Xiong et al., 1999).

Figure 1 shows the total volatile nitrogen value for four trademark of canned meat. No significant differences between the values of total volatile nitrogen for canned meat samples at a level (p < 0.05) and these values range from (12.1 to 12.7) mg N/100 g meat. These results were within the limits allowable of Iraq and the international specification (the Central Agency for Standardization and Quality Control, 1987), while the free nitrogen from proteins in canned meat does not exist and chemical changes as well were non-existent because the canned meat have detected components of salt and

**Table 3.** Lipid oxidation evaluation for four trademarks.

Trademarks	Free fatty acids %	Peroxide value (meq oxygen/kg lipid)	Thiobarbituric acid (mg malonldehyde/kg lipid)
Α	0.05±0.01 <sup>a</sup>	0.95±0.05 <sup>a</sup>	0.39±0.02 <sup>a</sup>
В	0.03±0.01 <sup>a</sup>	$0.45\pm0.05^{\circ}$	0.37±0.08 <sup>a</sup>
С	0.04±0.01 <sup>a</sup>	$0.65 \pm 0.05^{bc}$	0.45±0.005 <sup>a</sup>
D	$0.07\pm0.005^{a}$	0.80±0.10 <sup>ab</sup>	0.43±0.01 <sup>a</sup>

Means having the same letter in the same sections are not significantly different at  $P \le 0.05$ .

Table 4. Microbial assessment for four trademarks.

Trademarks	Total aerobic bacteria	Coliform Bacteria	Proteolytic bacteria	Lipolytic bacteria	Bacillus	Clostridium
Α	0.00	0.00	0.00	0.00	0.00	0.00
В	0.00	0.00	0.00	0.00	0.00	0.00
С	0.00	0.00	0.00	0.00	0.00	0.00
D	0.00	0.00	0.00	0.00	0.00	0.00

Table 5. Sensory evaluation of four trademarks.

Trademarks	Tenderness	Juiciness	Flavor	Color	Overall acceptability
Α	4.25±0.25 <sup>a</sup>	2.5±0.28 <sup>ab</sup>	2.75±0.25 <sup>a</sup>	2.75±0.47 <sup>b</sup>	4.0±0.00 <sup>a</sup>
В	4.25±0.47 <sup>a</sup>	3.5±0.28 <sup>a</sup>	$3.5\pm0.50^{a}$	2.0±0.40 <sup>b</sup>	4.5±0.50 <sup>a</sup>
С	3.25±0.47 <sup>ab</sup>	2.75±0.49 <sup>ab</sup>	2.75±0.75 <sup>a</sup>	3.0±0.40 <sup>ab</sup>	4.25±0.47 <sup>a</sup>
D	2.50±0.64 <sup>b</sup>	1.75±0.47 <sup>b</sup>	2.5±0.50 <sup>a</sup>	4.0±0.00 <sup>a</sup>	4.0±0.75 <sup>a</sup>

Means having the same letter in the same sections are not significantly different at  $P \le 0.05$ .

nitrate which helps to prevent the meat inside the cans from spoilage.

Free fatty acid (FFA), peroxide value and thiobarbituric acid for all trademarks of canned chicken meat are shown in Table 3. There are not significant differences among trademarks (p ≤ 0.05). All inspects samples A, B, C and D recorded 0.05, 0.03, 0.04 and 0.07% FFA, respectively. These percentages were within the limits recommended by the Central Agency for Standardization and Quality Control (1987). The canned meat was acceptable if the percentage (FFA) was not more than 1.5%. Peroxide values (PV) for A, B, C and D trademarks were 0.95, 0.45, 0.65 and 0.80 meg oxygen/kg fat, respectively. Overall were acceptable, the reason for the decline is due to the addition of nitrate salts and ascorbate, and this reduced the value of PV in meat (Al-Obaidi, 2005; Richards et al., 1998). Thiobarbituric acid (TBA) values were not significant between all trademarks, and it was acceptable because these values were within the limits recommended by the Central Agency for Standardization and Quality Control (1987), and it is not more than 2 mg.

The microbiological evaluation of the four trademarks of inspects samples are shown in Table 4. No significant

difference (p < 0.05) was found in total aerobic bacteria, coliform bacteria, proteolytic bacteria, lipolytic bacteria, *Bacillus* and *Clostridium*. The reason for not having bacteria in samples indicates the proper preparation of this meat and correct canning, and possibly the addition of some preservatives to it, especially nitrates, which have an important role in reducing the growth of anaerobic bacteria and their inhibition, especially *Clostridium* (Al-obaidi, 2005). According to the results, the process of canning scientifically was done properly and the handling and transporting were correctly carried out, so we have no contamination or any means of indication of aerobic bacteria.

The results in Table 5 show there were significant differences (p < 0.05) in the sensory properties (tenderness, juiciness and color) among the four trademarks of canned chicken meat, while no significant differences (p < 0.05) in the overall acceptability of the four trademarks of canned chicken meat by consumer. A trademark scored between 2.5 to 4.25 for tenderness, juiciness, flavor, color and overall acceptability, and B trademarks character was a light in color mark, while C trademarks character was dryness and weakness for juiciness and flavor, respectively but acceptable in color. D trademark

scored between 1.75 and 4, and acceptable in overall acceptability. Al-Rubeii et al. (2000) observed significant differences for the effect of genetics on the tenderness, flavor and juiciness that agree with the studied results according to the different companies with different meat samples.

#### Conclusion

From the results of this study, there were significant differences in chemical analyses among all trademarks of canned chicken meat. There were no significant differences among all trademarks in contents of free fatty acids and thiobarbituric acid, but there were significant differences among all trademarks in contents of peroxide values, and the total volatile nitrogen values for all trademarks were non-significant. There were no significant differences in microbial tests among all trademarks that indicated there were no aerobic bacteria in any of these trademarks. Significant differences in the sensory properties among the four trademarks were observed, while there were no-significant differences in the overall acceptability of the four trademarks.

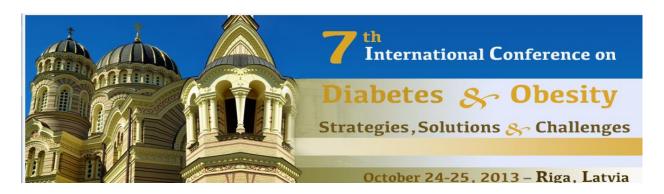
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