

Relation between serum Visfatin and clinical severity in different stages of rheumatoid arthritis

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Abstract: Background: Visfatin is a one of the recently discovered adipokine that has an important pro-inflammatory and catabolic roles in rheumatoid arthritis (RA). Pro-inflammatory cytokines, such as interleukin (IL)-1, IL-6, IL-15, IL-18 and tumor necrosis factor- α (TNF- α), initiate a number of physiological changes that result in the characteristic signs of inflammation. Since inflammation is the major factor leading to structural damage, it is critical to achieve rapid suppression of inflammation to maximize disease control. **Objective:** to evaluate the role of serum visfatin as a recent pro-inflammatory marker in RA according to the activity scores of disease to assess the possibility of introducing serum visfatin in the diagnosis and monitoring of RA patients and correlate between its serum level and other cytokines (IL-6 and TNF- α) and other laboratory biomarkers. **Patients and methods:** This study was conducted on a total number of 80 individuals, 60 of them were RA (48 females 80% , 12 males 20%) diagnosed as RA according to the American College of Rheumatology (ACR) / The European League Against Rheumatism (EULAR) 2010 criteria and 20 healthy subjects as a control (10 females 50%, 10 males 50%). RA patients were classified to 3 groups: Group I (severe RA): 20 RA patients ,Group II (moderate RA): 20 RA patients and Group III (mild RA): 20 RA patients according to clinical evaluation for disease activity assessed using a 28 joint disease activity score, (DAS-28). Blood samples were obtained from patients and controls for CBC and ESR. The sera of patients collected for ELISA estimation of serum visfatin , IL6 and TNF- α . CRP and RF were determined by turbidimetry quantitative method. **Results:** The comparison between the RA and control groups showed that; the mean serum level of visfatin, Platelets (PLT), ESR, IL6, CRP and RF were significantly higher in RA patients than control group . The comparison between the mild, moderate and severe RA groups showed that the mean levels of visfatin and IL-6 were significantly higher in severe RA group than moderate RA group which was significantly higher than mild RA group. There was a significant positive correlation between serum visfatin and {IL-6, ESR, CRP, TNF α and DAS 28} in RA group. **Conclusion:** Visfatin has a role in the pathogenesis of RA and could be considered as a disease marker in RA and a marker of radiographic bone damage and hence as a potential therapeutic target for RA. The finding of present study indicate also that serum visfatin and IL-6 might be of a valuable diagnostic value for RA, however the combined diagnosis using serum visfatin , IL-6 and RF test can improve RA diagnosis in early stage. Further studies are needed to determine the possibility of introducing visfatin as a potential therapeutic target especially in early RA to prevent erosions.

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1. Introduction

Rheumatoid arthritis (RA), the most severe of all joint diseases and also the most common systemic autoimmune disease, affecting approximately 1% of the adult population⁽¹⁾. The major features of RA are the activation and proliferation of synovial tissue and the degradation of articular cartilage. Synovial fibroblasts and inflammatory cells, such as macrophage, play key roles in this process. Innate immunity also plays an important role in the pathogenesis of RA⁽²⁾.

No single diagnostic test definitively confirms the diagnosis of rheumatoid arthritis. However, several tests can provide objective data that increase diagnostic certainty and allow disease progression to be followed⁽³⁾.

Failure to diagnose or treat a patient with RA at the early stages of the disease increases the risk of progression to persistent joint inflammation and damage. On the other hand, aggressively treating patients with mild arthritis, which probably will not evolve to erosive forms is also damaging. It exposes such patients to risk without proven benefits and represents the opposite of effective early treatment. Therefore, early diagnosis of those patients who will progress to more severe forms and consequently will require therapy that is more aggressive is essential⁽⁴⁾.

Visfatin was originally identified as a secreted growth factor for early lymphocytes (pre-B cell colony enhancing factor, PBEF). Visfatin or PBEF is a 52-kDa

protein found in living species from bacteria to humans (5).

Visfatin has shown both nuclear and cytoplasmic expression. Within the cell, it functions as a nicotinamide phosphoribosyl transferase, the rate-limiting step in a salvage pathway of nicotinamide adenine dinucleotide (NAD) biosynthesis. By virtue of this role, it can regulate cellular levels of NAD and so affect not only cellular energetics but also NAD-dependent enzymes such as sirtuins. It has been shown to be an adipokine expressed by fat cells and exerts a number of insulin mimetic effects (6).

Visfatin was added to a growing list of adipocytokines with potent effects on immunity and inflammation in addition to their metabolic activities (7). In CD14 (+) monocytes, visfatin induces the production of IL-1beta, TNF-alpha, and IL-6. Moreover, it increases the surface expression of co-stimulatory molecules CD54, CD40 and CD80 (8).

Rho et al., 2009, reported that visfatin is induced by inflammation and immune activation. It enhances B cell differentiation, initiation of cytokines and matrix metalloproteinases and inhibits neutrophil apoptosis thus playing a key role in persistence of inflammation (9).

Visfatin is released by a variety of cells and elevated levels can be found in the systemic circulation of patients with various acute and chronic inflammatory diseases including RA, sepsis, acute lung injury, inflammatory bowel disease and myocardial infarction (6).

A study on experimental animals proved that inhibition of visfatin lead to markedly reduced inflammation, arthritis severity and cartilage damage in a collagen-induced arthritis model. They postulated that pharmacologic inhibition of visfatin led to reduced levels of intracellular NAD in inflammatory cells and decreased production of TNF-alpha and IL-6 by such cells with clinical effects comparable to those of a TNF-alpha inhibitor in a murine arthritis model (10).

A large number of cytokines are active in the joints of patients with RA. It is now clear that these cytokines play a fundamental role in the processes that cause inflammation, particular destruction, and the comorbidities associated with RA. Following the success of TNF- α blocked as a treatment for RA, other cytokines now offer alternative targets for therapeutic intervention or might be useful as predictive biomarkers of disease. The biologic contribution and therapeutic potential of the major cytokine families to RA pathology, is focusing on (the TNF- α , IL-18, IL-6, IL-23, and IL-2 families) (11).

TNF- α is now recognized as mediating a wide variety of effector functions relevant to the pathogenesis of RA, including endothelial cell activation and chemokine amplification, leading to

leukocyte accumulation and probably attendant cardiovascular comorbidity; osteoclast and chondrocyte activation, promoting articular destruction; nociceptor sensitization; impaired cognitive function; and metabolic syndrome. These are all recognized components of the RA disease spectrum and explain the broad effects of TNF- α blockade in patients (12).

IL-6 is considered to play a central role in chronic inflammation and is expressed in excess at sites of inflammation. high levels of sIL-6R have been shown to correlate with the degree of joint destruction, in particular, in advanced stages of RA. IL-6 is a multitarget cytokine with activity relevant to RA. At the affected joints, IL-6 has a pivotal role in the inflammatory process, in osteoclast-mediated bone resorption, and in synovitis. IL-6 induces acute-phase proteins and contributes to the systemic manifestations of RA though hepcidin production (anemia) and acts potently in changing lipid concentrations (hypolipidemia). In addition, IL-6 may contribute to the induction and maintenance of the autoimmunity through B-cell activation and Th17 cell differentiation (13).

Aim of the study:

The present study aimed to evaluate the role of serum visfatin as a recent pro-inflammatory marker in RA according to the activity scores of disease to assess the possibility of introducing serum visfatin in the diagnosis and monitoring of RA patients and correlate between its serum level and other cytokines (IL-6 and TNF- α) and other laboratory biomarkers.

2. Patients and Methods

A total of 80 individuals consisted of 60 patients with the diagnosis of RA and 20 healthy subjects as a control are collected for this study. The study population was selected consecutively among patients who presented to the outpatient of Rheumatology clinics of Al-Hussein and Sayed Galal hospitals. The RA group Comprised 60 patients of age ranged from 18-60 years (mean 45y). They were 48 females and 12 males. Their disease duration ranged from one year to 23 years. Diagnosis of RA was based and confirmed according to (ACR)/ (EULAR) 2010 criteria (14).

RA patients were classified to 3 groups: Group I (severe RA): 20 RA patients with DAS28 range (5.22-7.32) (17 females and 3 males), Group II (moderate RA):20 RA patients with DAS28 range (4.00-4.96) (15 females and 5 males) and Group III (mild RA): 20 RA patients with DAS28 range (2.35-3.20) (16 females and 4 males). The control group comprised 20 whose age healthy subjects ranged from 24 to 57 years (mean 43y). They were 10 females and 10 males.

All patients with RA received prior medications. The drugs taken at the sampling time included 5mg of

prednisolone, methotrexate varying from 7.5-15mg/week, 200mg hydroxychloroquine and NSAIDs.

All patients were subjected to:

- 1- Complete history taking and full clinical examination with special attention to musculoskeletal system.
- 2- Disease activity was measured by disease activity score 28 (DAS28) in RA, consisting of a 28 tender joint count (range 0-28), a 28 swollen joint count (range 0-28), ESR, and an optional general health assessment on a visual analogue scale {range 0-100⁽¹⁵⁾}. The DAS28 has a continuous scale ranging from 0 to 9.4, and usually shows a Gaussian distribution in RA populations. The level of disease activity can be interpreted as low (DAS28 \leq 3.2), moderate (3.2 < DAS28 \leq 5.1), or high (DAS28 > 5.1). A DAS28 < 1.2 corresponds to being in remission according to the ARA criteria⁽¹⁶⁾.
- 3- Weight and height were measured for each subject then the body mass index was calculated as following: BMI = Body weight in Kg / height in m²⁽¹⁷⁾.
- 4- Laboratory investigations:

Sample collection:

Five ml of blood were withdrawn from each patient into two tubes:

- 1- 2ml of blood were immediately citrated for complete blood count (CBC) by Coulter device and

erythrocyte sedimentation rate determination (ESR) by westergren method.

- 2- 3ml of blood were allowed to be clotted, 1 hour later centrifuged for fifteen minutes and collected serum was stored at -30°C for determination of the following :
 - a) Serum visfatin was measured by Visfatin C terminal by a solid phase enzyme linked immunosorbent assay (ELISA) Kit (Phoenix Pharmaceuticals, Inc, Burlingame, CA, USA),⁽⁷⁾.
 - b) Serum CRP: by turbidimetry quantitative determination⁽¹⁸⁾.
 - c) Serum RF: by turbidimetry for quantitative determination⁽¹⁹⁾.
 - d) Serum TNF- α had been carried out using a solid phase enzyme linked immunosorbent assay (ELISA) Kit⁽²⁰⁾.
 - e) Serum IL-6: by a solid phase enzyme linked immunosorbent assay (ELISA) Kit⁽²¹⁾.

Statistical Methods:

Graph Pad Prism program version 5.0 was used for analysis of data. Data were summarized as mean \pm SD. One-way analysis of variance was used for analysis of more than two variables, followed by the Turkey's post-hoc test for the detection of significance. Simple linear correlation (Pearson's correlation) was also carried out. P-value of up to 0.05 was considered significant,⁽²²⁾ Oxford: Blackwell Science.

3. Results

Table (1): Mean \pm SD of demographic data and CBC in RA and control groups.

Variables	RA (N = 60)	Controls (N = 20)	P- value
Age (years)	45.1 \pm 11.8	42.6 \pm 8.5	0.831
BMI (kg/m ²)	22.26 \pm 3.55	23.1 \pm 3	0.670
WBCs (K/ μ l)	9.28 \pm 5.75	7.7 \pm 1.8	0.106
RBCs (K/ μ l)	4.72 \pm 0.62	5.0 \pm 0.48	0.241
Hb (g/dl)	11.95 \pm 1.54	12.33 \pm 1.47	0.987
PLT (μ l)	381.25 \pm 171.85	203.85 \pm 39.87	0.0001

The mean of PLT was significantly higher in RA patients than control group, while there was no significant difference in age, BMI, WBCs, RBCs and Hb between RA patients and control group.

Table (2): Mean \pm SD of laboratory data in RA and control groups.

Variables	RA (N = 60)	Controls (N = 20)	P- value
ESR (mm/hr)	43.75 \pm 24.9	14.25 \pm 6.65	0.0001
CRP (mg/L)	37.26 \pm 20.3	1.67 \pm 0.83	0.0001
RF (IU/ml)	91.8 \pm 83.0	0.61 \pm 0.24	0.0001
Serum visfatin (ng/ml)	18 \pm 5.14	4 \pm 1.2	0.0001
IL6 (pg/ml)	47.0 \pm 14.8	2.8 \pm 0.9	0.0001
TNF- α (pg/ml)	78.63 \pm 31.6	48.88 \pm 16.9	0.108

The mean ESR, CRP, RF, serum visfatin and IL- 6 were significantly higher in RA patients than control group. While, there was no significant difference in TNF- α between RA patients and control group.

Table (3): Mean \pm SD of laboratory data of rheumatoid arthritis patients in relation to severity of disease.

Groups Variables	Severe RA (N = 20)	Moderate RA (N = 20)	Mild RA (N = 20)	P- value
ESR (mm/hr)	45.19 \pm 25.58	46.63 \pm 25.65	37.53 \pm 23.391	0.536
CRP (mg/L)	50.33 \pm 18.09 ^a	34.88 \pm 16.89 ^b	16.77 \pm 12.84 ^c	0.0001
RF (IU/ml)	115.49 \pm 76.87	79.13 \pm 53.09	65.26 \pm 51.32	0.124
Serum visfatin (ng/ml)	22 \pm 7.21 ^a	17 \pm 4.91 ^b	9 \pm 2.14 ^c	0.0001
IL6 (pg/ml)	71.16 \pm 36.00 ^a	34.56 \pm 23.32 ^b	19.56 \pm 16.12 ^c	0.0001
TNF- α (pg/ml)	61.9 \pm 31.62	97.99 \pm 35.86	84.20 \pm 36.52	0.771

Different symbol indicates significance

The mean levels of CRP, serum visfatin and IL-6 were significantly higher in severe RA group than moderate RA group which were significantly higher than mild RA group. There was no significant difference in ESR, RF and TNF- α between the studied groups.

Table (4): Correlation between serum visfatin and other studied parameters in RA group.

Variable	Serum Visfatin(ng/ml)in RA group	
	r-value	p-value
Age (years)	0.208	0.152
BMI (kg/m ²)	0.285	0.134
Duration of disease (years)	0.328	0.074
ESR (mm/hr)	0.42	0.01
CRP (mg/L)	0.49	0.001
IL-6 (pg/ml)	0.51	0.0001
TNF- α (pg/ml)	0.44	0.001
DAS28	0.57	0.0001
VAS pain score	0.49	0.001

P value \leq 0.05 is considered to be significant.

There was a significant positive correlation between serum visfatin and ESR, CRP, IL-6, TNF- α , DAS28 and VAS pain score in RA patients. While, there was no significant correlation between serum visfatin and age, duration of disease and BMI in RA patients.

4. Discussion

Rheumatoid arthritis is the most common autoimmune chronic inflammatory arthritis, whose main characteristic is persistent joint inflammation that results in joint damage and loss of function⁽²³⁾. Early diagnosis of RA is an important challenge for clinical rheumatologists. This is because there is substantial evidence that early treatment with disease modifying anti-rheumatic drugs leads to a better disease outcome. As current predictors of joint destruction in RA have low specificity, serological biomarkers reflecting bone and cartilage destruction have been proposed as tools in assessing prognosis of this disease⁽²⁴⁾.

Cytokines regulate a broad range of inflammatory processes that are implicated in the pathogenesis of rheumatoid arthritis. In rheumatoid joints, it is well known that an imbalance between pro- and anti-inflammatory cytokine activities favors the induction of autoimmunity, chronic inflammation and thereby joint damage⁽²⁵⁾.

High levels of visfatin in plasma and synovial fluid have been found in RA patients⁽²⁶⁾. In the last few years, visfatin has generated much interest concerning its role during the development of RA⁽⁹⁾.

Since, the present study aimed to evaluate the role of serum visfatin as a recent pro-inflammatory marker in RA according to the activity scores of disease to assess the possibility of introducing serum visfatin in the diagnosis and monitoring of RA patients and correlate between its serum level and other cytokines (IL-6 and TNF- α) and other laboratory biomarkers.

In the present study, the comparison between the RA and control groups showed that, the mean of serum level of PLT was significantly higher in RA group than the control group (table 1).

This was data agreed with **Gülsüm, 2008, Jian et al., 2008 and Gasparyan et al., 2010** who had found that PLT was significantly higher in RA group than the control group and was increased by severity of RA disease^{(27) (28) (29)}.

In the present study the female percentage was 80 % in RA group. This data was agreed with the fact that women are affected by RA approximately 3 times more often than men⁽³⁰⁾.

Visfatin was reported to be an adipocytokine having proinflammatory and immunomodulating properties. However, the pathological role and clinical

relevance of visfatin in the setting of RA are still unclear⁽³¹⁾.

In this study, serum visfatin levels in RA patients were significantly higher than controls (table 2), and were significantly higher in severe RA group than moderate RA group which was significantly higher than mild RA group (table 3).

These findings go in hand with **Otero et al. 2006**, who investigated plasma levels of adipocytokines (leptin, adiponectin, visfatin and resistin) in patients with RA in comparison to levels estimated in healthy controls, and found patients with RA showed considerably higher plasma levels of leptin, adiponectin and visfatin than healthy controls, but no marked difference was observed in resistin levels between both Patients and controls⁽³²⁾.

Moreover, further research added that this adipocytokine was up regulated in synovial fluid of RA patients and demonstrated synovial fibroblasts as the major visfatin-producing cells in the rheumatoid synovium⁽³³⁾⁽³¹⁾.

Recently, **Senolt et al. 2011**, showed that serum visfatin levels were significantly higher in patients with RA compared with healthy controls and significantly decreased following treatment with anti- B cell therapy⁽³⁴⁾.

As inflammation plays a critical part in the pathophysiology of RA, the measurement of the acute-phase response is used as a surrogate marker of inflammation. The acute-phase reactants, CRP and ESR, are easy to perform, routinely available and the most widely used biological markers for assessing disease activity and inflammation in RA⁽³⁵⁾.

In the present study the mean serum level of ESR was significantly higher in RA group than the control group (Table 2).

This was also in agreement with **Jian et al., 2008, and Nalesnik, 2011**, who had found that the level of ESR in active RA patients was higher than that in normal control group⁽²⁷⁾⁽³⁶⁾.

In the current study the mean serum level of CRP was significantly higher in the RA group than the control group (table 2), and mean serum level of CRP in severe RA group was significantly higher than in moderate RA which was significantly higher than in mild RA (Table 3).

These results were supported with **Al-Mesry et al., 2003**, who had found CRP is a protein produced by the liver in response to tissue injury, infection and inflammation⁽³⁷⁾.

Also **Morovic-Vergles, 2008**, showed that serum CRP level was higher in RA and reflected a higher inflammatory activity in RA and CRP level increase by increasing of disease activity in RA patients⁽³⁸⁾.

Rheumatoid Factor RF is a very old serological marker for diagnosis of RA. RF is taken as a non-

specific marker of RA because it is also seen in other collagen vascular disease⁽³⁹⁾.

In the present study RF was significantly higher in RA than in the control group. This data was agreed with **Novikov et al., 2008**, who had found a variety of extra-articular features are typical of RA and it associated with the presence of rheumatoid factor in the serum⁽⁴⁰⁾.

The results were supported with **Khalifa and Abdelfattah, 2008, and Hui et al. 2010**, who found that there was a difference in RF level between RA group and non- RA groups⁽⁴¹⁾⁽⁴²⁾.

IL-6 is a multifunctional cytokine that regulates immune response and induces acute phase response. Despite the important physiological activities of IL-6, deregulated overproduction of IL-6 is pathologically involved in various immune-mediated inflammatory diseases including RA⁽⁴³⁾.

In the present study serum level of IL-6 was significantly higher in RA than in the control group and the level of IL-6 in the severe RA group was significantly higher than in the moderate RA group which was significantly higher than in the mild RA group.

These results were supported with **Fonseca et al., 2009**, as they found that IL-6 is a cytokine that can facilitate autoimmune phenomena, amplify acute inflammation and promote the evolution into a chronic inflammatory state in RA patients also **Cronstein, 2007**, had found that IL-6 is an important cytokine, present at elevated levels in patients with rheumatoid arthritis. The biological activities of IL-6 contribute to both systemic and local RA symptoms⁽⁴⁴⁾⁽⁴⁵⁾.

Also **Katz, 2001 and Sue-Yun Hwang, 2004**, had found IL-17, IL-1 β and TNF- α can induce IL-6 production according to disease activity⁽⁴⁶⁾⁽⁴⁷⁾.

The disease activity score is widely used to quantify disease activity and gauge response to treatment⁽¹⁶⁾.

Disease severity assessment relied on evaluation of pain using joint affection score (DAS 28) and laboratory evaluation of ESR and CRP levels. Such combination helped for patients' selection and goes in hand with **Klarenbeek et al., 2011**, who compared nine disease activity indices versus the American College of Rheumatology/European League against Rheumatism remission criteria in RA and tried to relate these indices to physical function and joint damage progression and found clinical DAS and simplified DI were the most stringent definitions of remission, DAS28 and DAS28-CRP had the highest proportions of remission and concluded that all indices, higher levels of disease activity were associated with decreased physical functioning and more radiological damage progression⁽⁴⁸⁾.

To focus more spot light on the relation of visfatin with damage of joints and other inflammatory markers,

the current study showed that there was positive correlation between serum visfatin and DAS28 in RA patients (Table 4).

These finding came in agreement with the findings reported by **Rho et al., 2009**, that, the visfatin concentrations were associated with higher Larsen scores, and this association remained significant after adjustment for age, race, sex, disease duration, BMI, and inflammation⁽⁹⁾.

These findings suggest a role for visfatin as a mediator of joint damage in RA. Such results are supported by experimental animal research that suggested that visfatin modulated inflammatory responses and radiographic joint damage in animal models⁽¹⁰⁾.

The current study showed also that there was positive correlation between serum visfatin and IL-6 and TNF alpha, in RA patients (table 4).

Recently, **Klein-Wieringa et al., 2011**, found that the levels of IL-6, TNF- α , visfatin, and adiponectin were positively associated with radiographic progression over 4 years and this association was independent of BMI and concluded that adipokines are predictors of radiographic progression in RA⁽⁴⁹⁾.

Additionally, **Nowell et al., 2006**,⁽³³⁾ reported that the production of visfatin was significantly up-regulated by IL-6 in human synovial fibroblast cell lines by a pathway dependent on signal transducer and activator of transcription 3 (STAT -3).

The current study showed also that there was no significant correlation between serum visfatin and age, duration of disease and BMI in RA patients (table 4).

These findings came in agreement with **Gonzalez-Gay et al., 2010**, and **Senolt et al., 2011**, found that serum levels of visfatin did not correlate with (BMI), age and duration of disease in patients with active RA in contrast to the case in non-RA subjects⁽⁵⁰⁾ ⁽³⁴⁾. This denotes that visfatin is an adipocytokine whose production in RA patients related principally to the disease process as part of the systemic inflammation and bone destruction suggesting a role for visfatin in the pathogenesis of RA.

The current study showed also that there was positive correlation between serum visfatin and both CRP and ESR, in RA patients (Table 4).

These findings came in agreement with another study that reported that visfatin was correlated with serum markers of inflammation (CRP, ESR) as well as clinical disease activity scores⁽³¹⁾.

The mechanism by which visfatin plays a destructive role in joints of RA patients is through activation of the transcription factors NF-kB and activator protein 1 and induction of IL-6, IL-8, MMP-1 and MMP-3 in RA synovial fibroblasts (RASFs) as well as IL-6 and TNF alpha in monocytes of these patients⁽⁵¹⁾.

These findings indicate that visfatin has a catabolic function in cartilage and may have an important role in the pathophysiology of arthritis. There is an evidence for an important function of innate immunity in the pathogenesis of RA⁽³¹⁾. While a study by **Luk et al., 2008**, demonstrated that visfatin plays a role as a novel mediator of innate immunity⁽⁶⁾. Taken together, these findings suggest that visfatin is involved in pro-inflammatory activity, innate immunity and cartilage-catabolic functions in the processes of RA⁽²⁶⁾.

Conclusion:

Visfatin has a role in the pathogenesis of RA and could be considered as a disease marker in RA and a marker of joint damage and hence as a potential therapeutic target for RA.

The finding of present study indicate also that serum visfatin and IL-6 might be of a valuable diagnostic value for RA, however the combined diagnosis using serum visfatin, IL-6 and RF test can improve RA diagnosis in early stage. Further studies are needed to determine the possibility of introducing visfatin as a potential therapeutic target especially in early RA to prevent erosions.

References

1. **Hoffmann M, Hayer S, and Steiner G (2009):** Immunopathogenesis of rheumatoid arthritis. *Ann N Y Acad Sci.*, 1173: 391-400.
2. **Brentano F, Kyburz D and Schorr O (2005):** The role of Toll-like receptor signaling in the pathogenesis of arthritis. *Cell Immune*; 233: 90 - 96.
3. **Moreland LW and Bridges SL (2001):** Early rheumatoid arthritis: a medical emergency? *Is; J Med.*, 111(6): 498-500.
4. **Mota LM, Laurindo IM and Santos Neto LL (2010):** Early rheumatoid arthritis: concepts. *Rev Assoc Med Bras*; 56(2): 227-9.
5. **Matsui H, Tsutsumi A, Sugihara M, Suzuki T, Iwanami K, Kohno M, Goto D, Matsumoto I, Ito S and Sumida T (2008):** Visfatin (pre-B cell colonyenhancing factor) gene expression in patients with rheumatoid arthritis. *Ann Rheum Dis.*; 67(4):571-2.
6. **Luk T, Malam Z, and Marshall J. (2008):** Pre-B cell colony enhancing factor (PBEF)/visfatin: a novel media innate immunity. *J Leucoc Biol.*; 83(4): 804-16.
7. **Fukuhara A, Matsuda M and Nishizawa M, (2005):** Visfatin: a protein secreted by visceral fat that mimics the effects of insulin. *Science*; 307: 426-30.
8. **Moschen AR, Kaser A and Enrich B (2007):** Visfatin, an adipocytokine with proinflammatory

- and immunomodulating properties. *J Immunol.* 178(3): 1748-58.
9. **Rho YH, Solus J and Sokka T, (2009):** Adipocytokines are associated with radiographic joint damage in rheumatoid arthritis. *Arthritis Rheum.* 60(7): 1906-14.
 10. **Busso N, Karababa M and Nobile M, (2008):** Pharmacological inhibition of nicotinamide phosphoribosyl-transferase/visfatin enzymatic activity identifies a new inflammatory pathway linked to NAD. *Plos One*; 3: 2267.
 11. **Fionula H (2008):** Evidence that cytokines plays a role in rheumatoid arthritis *J Clin Invest.* 3; 118 (11): 3537-3545.
 12. **Brennan FM and McInnes IB (2008):** Evidence that cytokines play a role in rheumatoid arthritis; *J Clin Invest*; 118(11):3537–3545.
 13. **Hashizume M and Mihara M, (2011):** The Roles of Interleukin-6 in the Pathogenesis of Rheumatoid Arthritis, Volume 2011 (2011), Article ID 765624, 8 pages.
 14. **Aletaha D, Neogi T, Silman AJ, Funovits J, Felson DT, Bingham CO 3rd, et al. (2010):** 2010 Rheumatoid arthritis classification criteria: An American College of Rheumatology/European League Against Rheumatism collaborative initiative. *Arthritis & Rheumatism.*, 62(9):2569–2581.
 15. **Scott J and Huskisson EC (1976):** Graphic representation of pain. *Pain*; 2: 175-84.
 16. **Fransen J and Van Riel CM (2005):** The DAS and the EULAR response criteria. *Clin Exp Rheumatol.*, 23 (39): S93-S99.
 17. **Bray, G.A. (1987):** Overweight is risk factor: definition, classification, prevalence, and risk. *Ann. N. Y. Acad. Sci.*, 499: 14 – 28.
 18. **Otsuji S, Shibata H and Umeda M (1982):** Turbidimetric immunoassay of serum C-reactive protein. *Clin Chem.*, 28: 2121-2124.
 19. **Winkles JW, Lunec J and Gray L (1989):** Automated enhanced latex agglutination assay for RF in serum. *Clin Chem.*, 35: 303-307.
 20. **Mesri, M (1998):** Endothelial cell activation by leukocyte microparticles. *J.Immunol.*161:4382-4387.
 21. **Grimsholm O, Rantapää-Dahlqvist S and Forsgren S (2008):** Levels of gastrin-releasing peptide and substance P in synovial fluid and serum correlate with levels of cytokines in rheumatoid arthritis; *Arthritis Research & Therapy* 2008, 10:410.
 22. **Armitage P, Berry G and Matthews JNS (2002):** *Statistical Methods in Medical Research* (4th edition). Oxford: Blackwell Science.
 23. **Carrasco R and Barton A, (2010):** Biomarkers of outcome in rheumatoid arthritis. *Rheumatology Reports*, 2(e3): 26-38.
 24. **Visser (2005):** Optimal dosage and route of administration of metho-trexate in rheumatoid arthritis. *Ann Rheum*; 68 (7): 94-99.
 25. **Iain B McInnes and Georg Schett, (2007):** Cytokines in the pathogenesis of RA. *Immunology*; 7, 429-442.
 26. **Bao JP, Chen WP and Wu LD (2009):** Visfatin; a Potential therapeutic target for rheumatoid arthritis. *The journal of International Medical Research*; 37: 1655- 1661.
 27. **Gülsüm S, (2008):** Increased platelet activation markers in rheumatoid arthritis are they related with subclinical atherosclerosis? *Informa*; 19 (2) 146-154.
 28. **Jian LIU, Rui-kai Z, Xue-fang YU, Yuan W (2008):** Effects of Xinfeng Capsule on platelet parameters, p-select, platelet ultra-structure and its therapeutic effect on rheumatoid arthritis patients in active phase. *China Journal of Traditional Chinese Medicine and Pharmacy*, 12-16.
 29. **Gasparyan AY, Stavropoulos-Kalinoglou A, Mikhailidis DP, Toms TE, Douglas KMJ and Kitas GD (2010):** Platelet functions in rheumatoid arthritis: arthritic and cardiovascular implications. *Rheumatology International*, 31(2):153-164.
 30. **Areskoug ED (2009):** A literature review of the sexual health of women with RA. *Musculoskeletal Care*; 7: 219-226.
 31. **Brentano F, Schorr O and Ospelt C, (2007):** Pre-B cell colony-enhancing factor/visfatin, a new marker of inflammation in rheumatoid arthritis with proinflammatory and matrix-degrading activities. *Arthritis Rheum.* 56(9): 2829-39.
 32. **Otero M, Lago R, Gomez R, Lago F, Dieguez C, Gómez-Reino JJ, and Gualillo O (2006):** Changes in plasma levels of fat-derived hormones adiponectin, leptin, resistin and visfatin in patients with rheumatoid arthritis; *Ann Rheum Dis.* 2006 September; 65(9): 1198–1201.
 33. **Nowell M, Richards P and Fielding C (2006):** Regulation of pre-B cell colony enhancing factor by STAT 3-dependent interleukin- 6 Trans signaling: implications in the pathogenesis of rheumatoid arthritis. *Arthritis Rheum.* 54; 2084-95.
 34. **Senolt L, Kryštůfková O and Hulejová H (2011):** The level of serum visfatin (PBEF) is associated with total number of B cells in patients with rheumatoid arthritis and decreases following B cell depletion therapy. *Cytokine*; 55(1): 116-21.
 35. **Nam J, Villeneuve E and Emery (2009):** The role of biomarkers in the management of patients

- with RA. *Current Rheumatology Reports*; 11: 371-377.
36. **Nalesnik M, Nikolić JM and Jandrić S (2011):** ADA and CRP in diagnosing and monitoring of RA. *Medicinski Glasnik*, 8(1): 163-168.
 37. **Al-mesry MR, Attwa ET and Omar HM (2003):** Assessment of cardiovascular risk factors in RA and OA. *Egypt Rheumatol Rehab*; 30(3): 323-332.
 38. **Morovic-Vergles J, Culo MI, Gamulin S and Culo F (2008):** Cyclic adenosine 5'-monophosphate in synovial fluid of rheumatoid arthritis and osteoarthritis patients. *Rheumatol Int.*, 29: 167-171.
 39. **Singh (2010):** Is rheumatoid factor still a superior test for the diagnosis of RA? *Rheumatol Int*; 30: 1115-1119.
 40. **Novikov AA, Aleksandrova EN, Karateev DE, Luchikhina EL, Demidova NV, Cherkasova MV, Denisov LN, and Nasonov EL. (2008):** Diagnostic value of antibodies to modified citrullinized vimentin in early rheumatoid arthritis, *Klin Lab Diagn.* 2008 Aug;(8):27-9.
 41. **Khalifa AIM and Abdelfattah A (2008):** Anti CCP2 and Anti Keratin Antibodies in patients with RA & OA. *The Egyptian Society of Rheumatology & Rehabilitation*; 35(1): 1-10.
 42. **Hui L, Wuqi S and Yang L (2010):** Diagnostic value of anti-CCP antibodies in northern Chinese Han patients with RA and its correlation with disease activity. *Clin Rheumatol.*, 29: 413-417.
 43. **Murakami, Miho, Nishimoto and Norihiro (2011):** The value of blocking IL-6 outside of rheumatoid arthritis: current perspective. *Current Opinion in Rheumatology*; 23 (3): 273-277.
 44. **Fonseca JE, Santos MJ H, Canhão H, Choy E (2009):** Interleukin-6 as a key player in systemic inflammation and joint destruction. *Autoimmunity Rev*; 8 (7): 538-542.
 45. **Cronstein BN (2007):** Interleukin-6-a key mediator of systemic and local symptoms in rheumatoid arthritis. *Bull NYU Hosp Jt Dis*; 65: 11-15.
 46. **Katz Y, (2001):** Interleukin-17 enhances tumor necrosis factor induced synthesis of interleukins 1, 6, and 8 in skin and synovial fibroblasts. *Arthritis Rheum*; 44:2176-2184.
 47. **Sue-Yun Hwang1, (2004):** IL-17 induces production of IL-6 and IL-8 in rheumatoid arthritis synovial fibroblasts via NF-kB- and PI3-kinase/Akt-dependent pathways. *Arthritis Res Ther*; 6:R120-R128.
 48. **Klarenbeek NB, Koevoets R, van der Heijde DM, Gerards AH, Ten Wolde S, Kerstens PJ, et al., (2011):** Association with joint damage and physical functioning of nine composite indices and the 2011 ACR/EULAR remission criteria in rheumatoid arthritis. *Ann Rheum Dis.* 70 (10):1815-21.
 49. **Klein-Wieringa IR, van der Linden MP, Knevel R and Kwekkeboom JC, (2011):** Baseline serum adipokine levels predict radiographic progression in early rheumatoid arthritis. *Arthritis Rheum.* ; 63(9):2567-74.
 50. **Gonzalez-Gay M, Vazquez-Rodriguez T and Garcia-Unzueta M (2010):** Visfatin is not associated with inflammation or metabolic syndrome in patients with severe rheumatoid arthritis undergoing anti-TNF-a therapy. *Clin Exp Rheumatol*; 28: 56-62.
 51. **Gosset M, Berenbaum F and Salvat C, (2008):** Crucial role of visfatin in matrix degradation and prostaglandin E2 synthesis in chondrocytes: possible influence on osteoarthritis; *Arthritis Rheum*; 58: 1399- 1409.

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