THE CLINICAL VALUE OF ANTI – CYCLIC CITRULLINATED PEPTIDE (ANTI- CCP) ANTIBODIES AND INSULIN RESISTANCE (IR) IN DETECTION OF EARLY AND SUBCLINICAL ATHEROSCLEROSIS IN RHEUMATOID ARTHRITIS

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ABSTRACT:

Aim of the work: To evaluate the role of anti-cyclic citrullinated peptide (anti ccp) antibodies**;** as an immunological marker which have been shown to be highly specific for RA and is predictor of disease severity**;** and also to evaluate the insulin resistance**;** as a key defect in the metabolic syndrome**;** for detection of early and sub-clinical atherosclerosis in rheumatoid arthritis patients. Also, to determine the specificity and sensitivity of anti-ccp and insulin resistance in evaluation of asymptomatic atherosclerotic RA patients.

Patients and methods: Fifty six (56) patients with rheumatoid arthritis were included in the present study. Nineteen (19) age and sex matched healthy subjects were recruited as controls for laboratory investigations and duplex ultrasonographic evaluation. All patients underwent a complete history review, clinical, laboratory, radiological examination, Duplex Ultrasonography of both carotids and Ultra-sonography to right brachial artery to detect endothelial function.

Results: There was a significant difference between patients and controls as regarding the mean, maximum, left IMT which was significantly higher in the first group ($p<0.05$). The overall prevalence of atherosclerosis in the studied patients was 30.37% all of them had thickened intima and only of them 5.4% had atherosclerotic plaque. Meanwhile, one (5.3%) of the controls had thickend intima with no detected plaque formation. The flow-mediated dilatation (endothelial dependent dilatation) in our study was significantly impaired in RA patients when compared to controls However, GTN responses did not differ between RA and controls.

A significant difference was found in HOMA-2 IR values ($p< 0.05$) between patient and control. Meanwhile, a statistically significant correlation was observed between thickened intima and flow mediated dilatation HOMA-2 IR values and a negative correlation between insulin resistance and post FMD, FMD dilatation percent and dilatation ratio as parameters which asses the endothelial function in RA patients.

There was association between anti-ccp and thickened intima media thickness and presence of plaques with a significant negative correlation between anti-ccp and endothelial function parameters in RA patients; FMD dilatation percent and dilatation ratio (0.005* and 0.001** respectively) and also it has a significant correlation with ultrasonographic duplex findings of carotid arteries.

Conclusions: There was significant association between IR and of subclinical atherosclerosis in our RA patients. Anti-CCP is a useful serological test for esta-blishing the diagnosis of rheumatoid arthritis and is associated with disease severity and we found association between anti-ccp and subclinical atherosclerosis in RA.

KEY WORDS:
Rheumatoid Arthritis Anti CCP Atherosclerosis
Insulin resistance Intima-media thickness.

INTRODUCTION:

Rheumatoid arthritis is a chronic inflammatory disease. Cardio-vascular events are the most important cause of mortality and morbidity in patients with rheumatoid arthritis (Ozbalkan et al., 2010). CVD mortality in RA appears to be predicted by the level of disease activity and severity of joint damage and extra-articular mani-festations. Some immunological markers, such as rheumatoid factor and anti-cyclic citrullinated peptide (anti-CCP) are more encountered in RA with extra-articular manifestations, anti-cyclic citrullinated peptide (anti-CCP) antibodies have been shown to be highly specific for RA and are more predictors, (even stronger) of disease severity and activity than rheumatoid factor. But until now; their association with CVD morbidity has not been examined by enough study **(**Gerli et al**.,** 2008).

Traditional risk factors which known to promote and accelerate the progression of atherosclerotic lesions are not common in patients with RA. There is other mechanisms play more effective roles in the increased cardiovascular complications of RA patients (Del Rincon et al., 2001, Pamuk et al., 2006 and La Montagna et al., 2007)

Insulin resistance is a pathogenetic factor in the metabolic syndrome and considers being the key defect in the metabolic syndrome. IR is associated with atherosclerotic risk factors which included in the metabolic syndrome like hypertension, hyperlipi-demia, and obesity that subsequently accelerate the development and progression of atherosclerosis; it was reported that there was peripheral IR associated with carotid as well as coronary artery atherosclerosis in RA patients and related to disease activity in absence of metabolic syndrome features; so impaired insulin sensitivity further predicts cardiovascular disease independent of other metabolic synd-rome features (Timar et al., 2000, Fujiwara et al., 2003 and Eckel et al., 2005) .

Therefore, outcome-based studies should be performed to assess the value of homeostasis model assess-ment of insulin resistance (HOMA-IR) [using the formula = (insulin (uU/ml) × glucose (mmol/l)) ÷ 22.5)] in RA, as an alternative and more sensitive approach should be devised to predict the cardiovascular risk in RA (Dessein et al., 2006, La Montagna et al., 2007 and Patrick et al., 2007).

Subclinical atherosclerosis can be demonstrated by an increased main carotid artery intima-media thickness (IMT), a good marker of generalized atherosclerosis. Measurement of caro-tid artery IMT is a noninvasive, sensi-tive, cost-effective method to deter-mine subclinical atherosclerosis and to diagnose at-risk patient groups (Park et al., 2002 and Del Rincon et al., 2003***).***

THE AIM OF THIS STUDY:

Is to evaluate the role of anti-cyclic citrullinated peptide (anti ccp) antibodies**;** as an immunological marker which have been shown to be highly specific for RA and is predictor of disease severity**;** and also to evaluate the insulin resistance**;** as a key defect in the metabolic syndrome**;** for detection of early and sub-clinical atherosclerosis in rheumatoid arthritis patients. Also, to determine the speci-ficity and sensitivity of anti-ccp and insulin resistance in evaluation of asymptomatic atherosclerotic RA patients.

PATIENTS AND METHODS:

Fifty six (56) patients with rheumatoid arthritis were included in the present study; (47) women and (9) men, their age ranged from (21-49 years) and disease duration ranged from six months to fifteen years. Nineteen (19) age and sex matched healthy subjects were recruited as cont-rols for laboratory investigations and duplex ultrasonographic evaluation.

Exclusion criteria:

Control and RA patients with known atherosclerotic complications such as stroke and MI; those undergoing hemodialysis; patients with peripheral vascular disease, malign-nancy, or infections; and hypertensive and diabetic patients were excluded.

All patients underwent a complete history review, clinical examination, laboratory (routine and special Laboratory investigation; Anti - cyclic citrullinated peptide Anti-CCP, Highly sensitive C- reactive protein (ELIZA) and Insulin and calculation of Insulin resistance IR by HOMA2-IR method), radiological examination (Anteroposterior radiographs of both hands and feet were done and scored by Simple Erosion Narrowing Score (SENS) was used for x-ray scoring**).**,

Duplex Ultrasonography of both carotids to determine the intimae media thickness (IMT) and to detect carotid plaques. The mean IMT (the mean of both right and left side) was assessed. At the same time the maximum IMT (the highest value either right or left) was also assessed. IMT is consider abnormal if > .072 cm and Ultrasonography to right brachial artery to detect endothelial function that FMD, GTN, and dilatation were calculated as follows:

FMD ={(Post FMD - Pre FMD)/Pre FMD}x 100.

GTN ={(Post GTN - Pre GTN)/Pre GTN}x 100.

Dilatation ratio= FMD/GTN.

STATISTICAL ANALYSIS

The statistical analysis was performed using SPSS 17.0. The following statistics were carried out:

The range, means and standard deviation were calculated for interval and ordinary variables and frequencies and percentages for categorical variables (Bland, 1987).

Comparisons were done by two procedures* Student's t-test: The independent samples T test was used to compare the means of two groups of cases. And***** the chi-squared (χ^2) test: The χ^2 is a nonparametric measure of the statistical independence of the categories of two variables measured on the nominal or dichotomous scale. We used the χ^2 test to test the significance of the differences between the two groups in categorical variables.

The Bivariate Correlations procedure computes Pearson's correlation coefficient with its significance levels. Pearson's correlation coefficient is a measure of linear association.

Sensitivity, specificity, positive and negative predictive values were calculated according to the their equations

RESULTS:

This study was carried out on 56 rheumatoid arthritis patients fulfilling the American College of Rheumatology (ACR) 1987 criteria for the classification of rheumatoid arthritis (Appendix A) and 19; age and sex matched healthy controls.

The demographic data of the RA patients and controls are shown in

table 1. And table 2 shows the Prevalence of insulin resistance and metabolic syndrome in rheumatoid arthritis patients and controls. A significant difference was found in HOMA-2 IR values (P = 0.005) between patient and control, whereas no significant difference emerged in the prevalence of metabolic syndrome or one or two of its factors.

Table 3 shows a comparison of parameters which asses the endothelial function between all the 56 RA patients and the 1 9 controls; Post FMD; (P= 0.0001), FMD dilatation percent (P = 0.0001) and dilatation ratio (p= 0.0001) were significantly lower in patient than controls.

Table 4 shows a comparison between 56 RA patients 19 controls with respect to ultrasonographic duplex findings of carotid arteries the mean, maximum and left IMT were significantly higher in patients compared to the controls.

Table 5 shows the overall prevalence of premature atheros-clerosis in RA patients and controls: in the first 17 patient has thickened IMT (30.36%) and only 3 patients (5.4 %) had atherosclerotic plaque; meanwhile, only one (5.3%) of controls had thickened intima with no detected plaque formation.

By comparing the laboratory measured parameters in patient and control groups as shown in table 6; the mean levels of Hg was significantly lower in the patient group than in control group, while the mean levels of ESR, US-CRP, RF, Anti-ccp, and HOMA28-IR were significantly higher in the patients group compared to control group.

Table 7 shows the anthropometric features, traditional cardiovascular risk factors, HOMA2-IR values and disease features in two groups divided according to the IMT more than 0.72 mm or equal or less than 0.72 mm. A significant difference were detected in both duration of the disease (P= 0.0001) and age (P= 0.01). Also there are significant difference in DAS28 (P= 0.02), Anti CCP (P= 0.001), Us-CRP (P = 0.0001) and HOMA2-IR (P = 0.003);

Table 8 shows negative correlation between insulin resistance and post FMD,FM D dilatation percent and dilatation ratio (0.005 and 0.01 respectively) as parameters which asses the endothelial function in RA patients. And significant correlation was fond with ultrasonographic duplex findings IMT of carotid arteries as shown in Table 9.

Table 10 There is association between Anti-ccp and thickened intima media thickness and presence of plaques with a significant negative correlation between Anti-ccp and endothelial function parameters in RA patients; FMD dilatation percent and dilatation ratio (0.005 and 0.001 respectively) and also it has a significant correlation with ultrasono-graphic duplex findings of carotid arteries as shown in table 11

As shown in Table 12 the sensitivity of anti ccp was less (94.12 %) than that of insulin resistance (100%) and its specificity was higher (46.15%) than that of IR (12.82%). And the positive predictive values of both of them were nearly equal, while the negative predictive value of IR was slightly greater.

Table (1):

		Patients (N = 56)	Control (N = 19)	P-value
Age	**Range**	21-49	22-49	0.350
	Mean±St.D.	36.50±7.22	34.6±8.22	
Sex	**Male**	9 (16.07%)	3 (15.8%)	0.977
	Female	47 (83.92%)	16(84.2%)	

Table (2):

	RA patien (n=56)	Controls (n=19)	P
Presence of insulin resistance (HOMA2–IR >1) n(%)	51of 56 patients (91.1%)	7 of 19 controls (36.8%)	0 .005
Presence of one feature of Metabolic syndrome, n (%)	27(48.2%)	10(52%)	0.647
Presence of two feature of Metabolic syndrome, n (%)	15(26.8%)	2(10.56%)	0.616
Presence of three features of Metabolic syndrome, n (%)	2(3.6%)	1(5.3%)	0.398

HOMA2-IR= Homeostasis model assessment, IR= insulin resistance.

Table 3:

		RA patients (n=56)	Controls (n=19)	P
Pre.fmd(average)	Mean±SD	3.69 ± 0.51	3.9 ± 0.49	0.121
Post.fmd(average)	Mean±SD	4.32 ± 0.60	5.11 ± 0.52	0.0001
Fmd dilatation %	Mean±SD	17.30 ± 7.72	28.52 ± 10.11	0.0001
Pre.GTN(average)	Mean±SD	3.71 ± 0.54	3.91 ± 0.49	0.150
post.GTN(average)	Mean±SD	4.63 ± 0.72	4.89 ± 0.62	0.131
GTN.dilatation %	Mean±SD	24.87 ± 9.02	25.72 ± 9.67	0.727
Dilatation ratio	Mean±SD	0.71 ± 0.261	1.078 ± 0.189	0.0001

FMD= flow mediated dilatation , GTN= glyceryltrinitrate.

Table (4): Comparison of Ultrasonographic duplex findings in RA patients and controls

		RA patients (n = 56)	Controls (n = 19)	P
Mean IMT	Mean ± SD	0.057 ± 0.016	0.050 ± 0.007	0.018
Lt. IMT	Mean ± SD	0.057 ± 0.016	0.049 ± 0.008	0.01
Rt. IMT	Mean ± SD	0.068 ± 0.087	0.0495 ± 0.0869	0.11
Maximum	Mean ± SD	0.0597 ± 0.017	0.0526 ± 0.008	0.02

IMT= intema media thickness

Table (5): Prevalence of premature atherosclerosis in RA patients and controls

	RA patients (n = 56)	**Controls (n = 19)**
Premature atheroscleros	Frequency	Frequency
Thickened intima	17 (30.36%)	1(5.3%)
Plaque	3 (5.4%)	0 (0%)

Table (6): Laboratory features of RA patients and controls:

	RA patient (n = 56)	**Controls (n =19)**	
	Mean±SD	**Mean±SD**	**P**
Hb(gm%)	11.82 ± 2.04	12.82±1.79	0.05
WBCs (/mm)	6.08 ± 2.28	7.46±2.74	0.5
Platelets (/mm)	286.68 ± 88.33	269.79±60.95	0.4
ESR (mm/hr)	40.82 ± 42.27	16.26±8,74	0.0001
FBS (mg%)	90.7 ± 17.43	88.42 ± 10.27	0.5
AlT	23.05 ± 11.61	21.37 ± 8.32	0.5
AsT	24.73 ± 21.14	24.37 ± 8.48	0.9
Total cholesterol(mg/dl)	166.25 ± 30.76	170.42 ± 36.65	0.63
Triglycerides(mg/dl)	90.70 ± 30.72	81.42 ± 26.8	0.22
HDL(mg/dl)	42.8 ± 3.4	42.37 ± 4.4	0.63
LDL(mg/dl)	114.45 ± 33.4 8	122.26 ± 35.41	0.406
RF	90.85 ± 203.48	0.87 ± 0.45	0.001
Anti ccp	85.11 ± 81.98	0.86 ± 0.46	0.0001
US –CRP	70.4 ± 61.71	0.54 ± 0.64	0.0001
HOMA2- IR	87.96 ± 270	5.34 ± 8.4	0.02

Hb= Hemoglobin, WBC= White Blood Cells, ESR= Erythrocyte Sedimentation Rate, FBS= Fasting Blood Sugare, AlT= Alanine transaminase, AsT= Aspertate transaminase , HOMA2-IR= Homeostasis model assessment, IR= insulin resistance, RF= rheumatoid factor.

Table (7): Comparison of RA patients features with an increased and normal mean IMT

		IMT ≤ 0.72mm N = 39	**IMT > 0.72mm N = 17**
Age	34.9 ± 6.9	40.12 ± 6.5	0.01
Disease duration	4.8 ± 3.06	8.8 ± 4.5	0.0001
DAS28	4.85 ± 1.35	5.8 ± 1.5	0.02
Obesity(BMI)	26.7 ± 5.4	24.5 ± 8.2	0.23
Systolic BP (mm/Hg)	117.23 ± 8.13	120.29 ± 10.38	0.37
Diastolic BP	76.81 ± 4.83	78.82 ± 5.16	0.21
Us-CRP	48.48 ± 51.599	120.79 ± 53.89	0.0001
IR	19.61 ± 43.5	244.75 ± 456.7	0.003
Cholesterol	162.15 ± 28.6	175.7 ± 34.4	0.132
Triglyceride	87.1 ± 30.6	98.8 ± 30.15	0.21
HDL	43.33 ± 4.4	42.85 ±3.3	0.07
LDL	110.62 ± 31.45	123.24 ±37.2	0.2
Cumulative steroids doses	748.21 ± 942.575	1335.88 ± 1621.5	0.005
Anti-ccp	61.3 ± 69.7	139.6 ± 83.9	0.001

IMT= intima media thickness, DAS28= disease activity score, BMI=body mass index, BP= blood pressure, Us-CRP=ultra sensitive c-reactive protein, IR= insulin resistance, HDL= high density lipoprotein, LDL= low density lipoprotein, Anti-ccp= anti cyclic citrullinated peptide.

Table (8): Correlation between insulin resistance and endothelial function in RA patients

Endothelial function	**IR**	
	(r)	P
Pre.fmd(average)	0.139	0.308
Post.fmd(average)	-0.091	0.04
Fmd dilatation %	-0.403	0.005
Pre.GTN(average)	0.141	0.23
post.GTN(average)	0.039	0.715
GTN dilatation %	0.123	0.301
Dilatation ratio	-0.112	0.012

(r)= Correlation coefficient, FMD= flow mediated dilatation, GTN= glyceryltrinitrate.

Table (9): Correlation between insulin resistance and Ultrasonographic duplex findings in RA patients

duplex findings	IR	
	(r)	P
Rt IMT	0.051	0.71
Lt. IMT	0.266	0.005
Mean IMT	0.344	0.009
Maximum	0.344	0.009

IMT= intema media thickness, (r)= Correlation coefficient

Table (10): Correlation between anti ccp and endothelial function in RA patients

Endothelial function	Anti-ccp	
	(r)	P
Pre.fmd(average)	0.275	0.23
Post.fmd(average)	0.224	0.02
Fmd.dilatation %	-0.091	0.005
Pre.GTN(average)	0.263	0.39
post.GTN(average)	0.171	0.72
GTN.dilatation %	0.294	0.31
Dilatation ratio	-0.437	0.001

(r)= Correlation coefficient, FMD= flow mediated dilatation, GTN= glyceryltrinitrate.

Table (11): Correlation between anti ccp and Ultrasonographic duplex findings in RA patients

duplex findings	Anti-ccp	
	(r)	P
Rt IMT	0.249	0.06
Lt. IMT	0.471	0.0001
Mean IMT	0.458	0.0001
Maximum	0.464	0.0001

IMT= intema media thickness, (r) = Correlation coefficient

Table (12): Sensitivity and specificity of anti-CCP and IR for presence of premature atherosclerosis in RA patients:

	Sensitivity (%)	Specificity (%)	PPV	NPV
Anti-CCP	94.12 %	46.15 %	0.34	0.94
IR	100 %	12.82%	0.33	1

Anti-ccp= anti cyclic citrullinated peptide**,** PPV= positive predictive value, NPV= negative predictive value.

DISCUSSION:

The included Fifty six (56) patients with rheumatoid arthritis were forty seven (47) females (83.92%) and nine (9) males (16.07%) and their age ranged from (21years) to (49 years); disease duration ranged from six months to fifteen years copared to 19 ages and sex matched healthy controls.

Our patients and controls were much younger (the patients's mean age was 36.50±7.22 years while the control's mean age was 37.2±10.97 years) than the patients and control studied by La Montagna et al., (2007) (mean age 53.8±11.6 and 51.8±11 years, respectively). The relatively early age of onset of our sample may attribute to methodological differences between the studies.

In that study of La Montagna et al., (2007) and similarly in our study both patients and control were investigated for their demographic characteristics, namely: sex, age and menopausal status.The median disease duration 6 (2-10) years and controls had a mean age of 40.3 ±11.6 years and 42.7±12.6 years respectively, with mean disease duration of 9..9± 8.7 years.

Both patients and controls were subjected to endothelial function assessment. In addition, right and left carotid arteries were evaluated for intima media thickness and for the presence of plaque using high resolution B-mode ultrasound.

In our study; the prevalence of traditional cardio vascular risk factors in rheumatoid arthritis patients and controls were of no significant difference was found between the two groups studied.

In our study, obesity, as defined by BMI > 30.0 was reported in 20(35.7 %) of our RA patients versus 6(31.6%) of our healthy controls with no significant statistical differences between both groups. Our results were similar to La Montagna et al., (2007) study, where there were no statistical significant differences between patients and controls.

Lipid profile in our RA patients were: total cholesterol ranged from 119-268 mg/dl with a mean of 166.25 ± 30.76, triglycerides ranged from 40-167mg/dI with a mean of 90.70+30.7, high density lipoprotein ranged from 34-50 mg/dl with a mean of 42.93±3.53 while low density lipoprotein ranged from 69-197 mg/dl with a mean of 114.45±33.43.

However, we did not find this association, in the present study this agrees with results of (Walberg-Jonsson et al., 1999 and La Montagna et al., 2007) studies. La Montagna et al., (2007) compared Forty-five patients with RA and 48 healthy controls, and found that RA patient had no significant raised triglycerides and LDL cholesterol levels, and significantly lower HDL cholesterol levels also we and La Montagna et al. 2007; did not detect significant differences between patients and controls in other metabolic risk factors; hypertension and increased BML Instead, Wallberg-Jonsson et al., (1999) found a significant association with BP (Wallberg-Jonsson et al., 1999).

The discrepancy in results is likely to depend on differences among the cohorts of patients investigated, including differences in their nutritional habits. Nevertheless traditional risk factors clearly play a role in the pathogenesis of atherosclerosis in RA and must be investigated when evaluating the individual RA patient (Cuomo et al., 2004).

Inflammation is considered to be an important risk factor for premature atherosclerosis in RA. This was a very consistent finding in the present study as measured by acute phase reactants including highly sensitive CRP and erythrocyte sedimentation rate.

As expected , higher levels of ESR ($p < 0.001$) and CRP($P < 0.001$) were detected in RA compared to healthy controls in agreement with Pereira et al., (2009) higher levels of ESR ($p < 0.001$) and CRP ($P < 0.001$) also were detected . These data definitely support systemic inflammation as a factor of importance for premature atherosclerosis in RA patients.

In this study there were significant correlation between US-crp and subclinical atherosclerosis markers; endothelial function [FMD dilatation percent and dilatation ratio (0.005* and 0.002** respectively)] and ultrasonographic duplex findings of carotid arteries in RA patients. These findings were similar to the results of Michael et al., (2010) who found also significant correlation between US-crp and subclinical atherosclerosis and saw that his study findings suggest that the use of CRP in RA (and perhaps other chronic inflammatory disease) should be further evaluated before used as a risk of CVD.

However, Elias - Smale Se et al., (2007) see that the carotid US findings in RA patients can not be predicted by isolated laboratorial parameters such as CRP or single time point disease activity assessment, in contrast to the normal population in which isolated acute phase tests are related to atherosclerotic findings (Elias- Smale et al., 2007) .

This finding suggests that these markers may lose their discriminatory power with respect to cardiovascular outcomes among patients with conditions in which levels of inflammatory molecules are elevated, such as RA, in contrast to the general population; where most values fall within the normal range. "Nevertheless, levels of these markers of inflammation were abnormal in many patients with RA and longitudinal, rather than cross-sectional, studies many yet define their relation to the development and progression of atherosclerosis.

The relationship between IR and subclinical atherosclerosis and regarding glucocorticoid was reported in many studies. The present study confirm these previous finding of an increased prevalence of insulin resistance in RA compared to controls ($p<0.001$), and indicate a significant correlation between HOMA2-IR and increased IMT both simple linear regression and in stepwise multiple regression with the latter, IMT was independently related to HOMA-2-IR, with steroid exposure, Mets, number of its components and diastolic BP acting as other contributors. On the whole, our data show, a relationship between subclinical atherosclerosis, insulin resistance and glucocorticoid use that is in agreement with the findings of other authors (Hanley et al., 2002, Kitas GTD et al., 2003, del Rincon et al., 2004, Dessein et al., 2005 and La Montagna et al., 2007).

Insulin resistance, a central component of MetS, has been shown to be an independent risk factor for ischemic heart disease and had long been known to occur in RA (Paolisse et al., 1991, Svenson et al., 1998 and Bonora et al., 2007).

In our study, a significant difference was -found in HOMA2-IR values ($p < 0.001$) between patients

and controls, whereas no difference emerged in the prevalence of Mets or one or two of its factors.in addition, no significant difference was found in the frequency of either MetS or abnormal values of H0MA2-1R in RA steroid users as compared to non-users. This in agreement with Pereira et al., (2007) results.

In La Montagna et al., (2007) study a significant correlation was also found between HOMA 2-IR and DAS28 ($r=0.549$, $p<0.001$), similarly in our study: also there is correlation was also found between HOMA2-IR and DAS28.

In this study, the relationship with DAS28, a composite disease activity index in RA, supports in role for chronic inflammation in the development of insulin resistance. The relationship between HOMA2-IR, 1MT, DAS28 and glucocorticoid cumulative doses confirms that IR in RA is associated with inflammation in agreement with La Montagna et al., (2007) results.

Some immunological markers, such as rheumatoid factor, antinuclear antibodies and anti-cyclic citrullinated peptide (anti-CCP) antibodies are more often encountered in RA with extra-articulare manifestation (Gerli et al., 2008).

The frequency major imunoinflamatory markers of RA in our series is also comparable with other reportes. RF was detected in the majority of RA patients (64.6%). This is in agreement with La Montagna et al., (2007*)*; results (69.6%) and in (83%) in Pereira et al., (2009) study . While in Pamuk et al., (2006) study; where sixty three patients with RA and 34 controls were studied; only 46% were RF positive.

Also in our study anti-ccp antibodies were positive in 47 patient (83.9) and ranged from 1-312 with mean of 85.11±81.98. While in Pereira et al., (2009*)* study anti-ccp antibodies were detected in 56 RA patienta (78.9%), and 52 of the 81 patients analyzed (64.2%) tested positive for anti-CCP antibodies in Gerli et al., (2008) study.

In our study there is association between anti ccp and thickened intima media thickness and presence of plaques with a significant negative correlation between anti-ccp and endothelial function parameters in RA patients; FMD dilatation percent and dilatation ratio (0.05* and 0.001**) respectively and also it has a significant correlation with ultrasonographic duplex findings of carotid arteries.

This in agreement with Gerli et al., (2008), who to our knowledge, was the first report showing an association between anti-CCP and subclinical atherosclerosis in patients with RA. The association of anti-CCP antibodies and CVD risk may simply reflect the effect of a more diffuse and aggressive disease, possibly explaining the apparently weaker association of these autoantibodies specifically with arterial wall damage.

Turesson et al., (2007) Conclude that Rheumatoid factor is strongly associated with severe ExRA manifestations in patients with rheumatoid arthritis, and similar but weaker associations exists for anti-CCPs and suggest a role for rheumatoid factor and anti-CCP in the pathogenesis of ExRA.

In contrast, in Pereira et al., (2009) study who found that although anti-cyclic citrullinated peptide (anti –

ccp) antibodies) have been shown to be significantly associated with RA patients group; but their association with thickened intima media thickness; as subclinical atherosclerosis parameter; were not of significant results.

And so Pereira et al., (2008) Conclude that there is a clear association between all autoantibodies studied ; include RF and Anti CCP; and increased IMT or presence of plaques was not observed. The great prevalence of carotid atherosclerosis in RA was related to age, total and LDL cholesterol, as identified in normal population.

Intima-media thickness (IMT) of carotid arteries was determined by B-mode ultrasonography, and presence of atheromatous plaques was determined. The presence of accelerated atherosclerosis is well documented in RA patients when compared to the general population.

In the present study there is an overall higher prevalence of premature atherosclerosis in RA patients than controls in the first; 17 patients has thickened IMT (30.36%) and only 5.4% (3 patients) had atherosclerotic plaque; while, no one of controls had thickened intima with no detected plaque formation. In addition, the mean IMT was significantly higher in RA patients than our healthy controls (0.57±_0.051 versus 0.49±0.072) mm respectively (P = 0.01*).

And by comparison between 56 RA patients 19 controls with respect to ultrasonographie duplex findings of carotid arteries and the endothelial function; as two markers of subclinical atherosclerosis; the mean right and left IMT were higher in patients compared to the controls.

The mean IMT was marginally in RA patients versus controls (0.560.52 vs .49 $\pm$ 0.7 mm, p= 0.05) and plaques were also more frequently observed in RA (5.4%vs. 0%, p=0.02). And by comparison of parameters which asses the endothelial function in RA patients and controls. Only FMD (P = 0.043) and dilatation ratio (p=0.00l) were significantly lower in patient than controls. These result in agreement of other studies of Pamuk et al., 2006, La Montagna et al., 2007 and Pereira et al., 2009.

In accordance with our result, a study by La Montagna et al., 2007***;*** who reported that 50% of patients with RA have atherosclerosis; mean IMT was 0.754±0.11mm. The 45 RA patients were, ranging from 0.56 to 0.99 mm. In the 48 control subjects, mean IMT was 0.659±0.093 mm ranging from 0.48 to 0.88 mm ($p< 0.001$) in addition, studies using a control group, also confirm that the prevalence of the disease is higher in RA patients than in the general population.

Epidemiological studies have used different out off values for IMT in the general population. Differences in the site and method of carotid measurement may account for different results.

Many studies have assessed atherosclerosis at different sites the within the carotid, and it is unclear to what extent the choice of site has influenced the results some studies focused on the common carotid IMT (El-Magadmi et al., 2004), others on the internal carotid IMT and still others on the average IMT in the common carotid artery, carotid bulb, and internal carotid artery (Doria et al., 2003), Thickening may progress at

different rates in the common carotid and internal carotid arteries, and associations with risk factors may differ between these sites.

Another important factor is that the cut off point between normal and high IMT was different between studies. In Doria et al., (2003) study normal IMT was defined when complex intima-media is < 0.09 cm; therefore, IMT values 0.09 cm were considered indicative of thickened intima. While in Marazini et al., (2005) study, the subclinical atherosclerosis IMT cut-off value delected was > 0.7 mm.

In our study intima-media thickness is considered abnormal if > 0.072 cm which is in agreement with study of (Denarie et al., 2000 and La Montagna et al., 2007)

Differences in the ultrasound equipment or even using the same equipment with different frequency or different resolution could result in such variability. Common carotid IMT using ultrasound with a high frequency may allow more accurate results than of limited resolution. Different scanning and reading protocols could also influence the results.

Another factor reported that IMT In men is significantly thicker than that in women when estimated using 7-8 MHz frequency ultrasound (Lawlor et al., 2004) difference in the methodology for assessment of atherosclerosis may lead to different results.

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الملخص العربى

مرض الروماتويد المفصلي هو مرض من أمراض خلل المناعة ، يصيب حوالى 5,.- 1 ٪ من سكان العالم وهو غير معروف السبب. يصيب مرض الروماتويد المفصلي أساساً المفاصل الصغيرة لليدين و القدمين و يؤدى إلى حدوث تشوهات بالمفاصل إذا لم يتم علاجه فضلا عن الاعرض او المضاعفات الغير مفصلية واهمها امراض القلب والاوعية الدموية ومن بينها التصلب الشريانى (العصيدى) والذى اثبتت الكثير من الأبحاث حدوثه بنسب اعلى لدى هؤلاء المرضى مما قد يؤدى بحياة المريض.

وحيث ان حدوث أضطرابات القلب والاوعية الدموية في حالات الروماتويد المفصلى من اخطر المضاعفات التى قد تؤدى بحياة المريض ، فأن التشخيص المبكر لتصلب الشرايين يحمى المريض من مشاكل صحية ضخمة كجلطات شرايين القلب والمخ .

ولذا فأنه من الضروري البحث عن الجديد فى التحاليل الطبية وغيرها من الوسائل التشخيصية للاكتشاف المبكر للمرض قبل ظهور أى أعراض أومضاعفات.

وقد وجد أنه بشكل فسيولوجى ومع التقدم العمرى يحدث زيادة تدريجية فى سمك طبقات الجدار الوعائى الى ان تصل الى الصورة المرضية وهى التصلب الشرايينى وهناك العديد من عوامل المخاطرة التقليدية مثل السمنة وارتفاع ضغط الدم وارتفاع السكر بالدم وارتفاع نسبة الكوليستيرول والدهون بالدم ذلك مما قد يؤثرعلى متلازمة التمثيل الغذائى مؤديا الى الاسراع بحدوث التصلب الشرايينى.

وفى حالات الروماتويد المفصلى هناك عوامل اخرى فضلا عن هذه العوامل تؤدى الى الاسراع بحدوث التصلب الشرايينى ومضاعفاته ومن اهم هذة العوامل هو الالتهاب كعملية مرضية تحدث ليست بالمفاصل فقط بل تمتد لتشمل الجدار الوعائى ومن هنا كان لتحديد شدة الالتهاب ونشاط المرض اهميته فى التوقع بسرعة حدوث التصلب الشرايينى وسرعة التدخل العلاجى لايقافة وعلاجه.

وحيث ان هناك العديد من الابحاث الطبية الحديثة تقر اهمية تحليل الانسولين رسيستانس كعامل غير تقليدى فى التأثيرعلى متلازمة التمثيل الغذائى مؤديا لاحداث التصلب الشرايينى. وكذلك فإن مضاد سى سى بى هو أحد الأجسام المضادة الذاتية التى اكتشفت حديثا فى مرضى الروماتويد المفصلي و قد أبرزت نتائج الأبحاث فائدته فى التشخيص المبكر لمرض الروماتويد المفصلي لما يتمتع به من حساسية و خصوصية عالية لهذا المرض, كما وجد أن له علاقة بنشاط المرض وشدته فى هؤلاء المرضى ويمكن التوقع بوجود ارتباط بينه وبين حدوث التصلب العصيدى المبكرعندهم.

ومن هنا جأت اهمية دراسة الأهمية الأكلينيكية لكل من الانتى سى سى بى والانسولين ريسيستانس كوسائل معملية سهلة للتوقع وجود التصلب الشرايينى فى حالات الروماتويد ومن ثم تشخيصه باستخدام الاشعة التلفيزيونية (الدوبلكس) بجهاز الموجات الفوق صوتية كوسيلة سهلة وغير مكلفة لمعرفة حدوث التصلب العصيدى من عدمه.

الهدف من الدراسة:

يهدف هذا البحث الى دراسة مدى الاهمية الاكلينيكية لمضاد البيبتيد السيتروليناتيد الحلقى والانسولين ريزيستانس فى التشخيص المبكر لتصلب الشرايين (التصلب العصيدى) فى حالات الروماتويد المفصلى.

وقد اجريت هذه الدراسة على ستة وخمسون مريضة بالروماتويد فى مقارنة مع تسعة عشر من الاصحاء من نفس السن والنوع وقد أجريت للمرضى دراسات عديدة وهى:

- تاريخ المرض.
- فحص اكلينيكى عام.
- فحص الجهاز الحركى.
- فحوصات معملية (صورة دم كاملة و سرعة ترسيب ودهون كاملة بالدم وسى ريأكتيف بروتين على الحساسية و معامل روماتويد و مضاد سى سى بى وانسولين ريزيستانس).
- أشعة سينية لليدين و القدمين.
- تشخيص التصلب العصيدى باستخدام الاشعة التلفيزيونية (الدوبلكس) بجهاز الموجات الفوق صوتية وقد قمنا بقياس سمك الطبقة المبطنة والطبقة المتوسطة للشريان السباتى الايمن والايسر , او وجود التكونات العصيدية لكل من المرضى والاصحاء. وايضا تم استخدام الاشعة التليفيزيونية فى فحص وظيفة الغشاء المبطن للاوعية الدموية عن طريق تحديد نسبة اتساع الشرايين نتيجة تدفق الدم فيها من بعد انسدادها ومن بعد اعطاء عقار النترات على الشريان العضدى الايمن.

وقد اجرى قياس لسمك الطبقة المبطنة والطبقة المتوسطة للشريان السباتى فى كل من المرضى والاصحاء ووجد ان الفارق ذو دلالة احصائية حيث وجد ان متوسط سمك الطبقة المبطنة والطبقة المتوسطة فى المرضى 0,06±0,01 ,بينما متوسط سمك الطبقة المبطنة والطبقة المتوسطة فى المجموعة السليمة 0,05 ±0,01.

وقد أظهرت النتائج أن هناك زيادة فى سمك الطبقة المبطنة والطبقة المتوسطة للشريان السباتى فى سبعة عشر مريضات بالروماتويد (30,36 %) مع وجود شرائح فى الشريان السباتى فى ثلاث منهم . بينما يوجد زيادة فى سمك الطبقة المبطنة والطبقة المتوسطة للشريان السباتى فى حالة واحدة (5,3 %)من الاصحاء .

وقد اظهرت النتائج انخفاض متوسط نسبة اتساع الشرايين نتيجة تدفق الدم فيها من بعد انسدادها فى المرضى عنه فى الاصحاء بفارق ذى دلالة احصائية.

وقد تم استنتاج وجود علاقة ذات دلالة احصائية بين مقاومةالانسولين ووجود التصلب العصيدى المبكر وكذلك وجود الاجسام المضادة الذاتية انتى سى سى بى.

ذلك مما يجعنا نوصى باستخدام تحديد نسبة كل منهما بالتحاليل المعملية كاختبار حساس ومتخصص للتوقع والاشتباة فى وجود التصلب العصيدى المبكر مما يستتبع "فى حال النتائج الايجابية" التوصية بعمل الفحص بالاشعة التليفيزيونية بالموجات الفوق صوتية (دوبليكس) لتشخيص المبكر للمرض والتحكم فية وعلاجه بالطرق العلاجية المختلفة بداية من علاج الروماتويد نفسه والوصول الى الاستقراراو الخمول المرضى التام مما يوقف تاثير عامل الالتهابات على ذيادة التصلب وكذلك تقليل نسب الكورتيزون المستخدم قدر الامكان نظرا لان حدوث المرض متناسب مع الجرعة التراكمية للعقار وايضا استخدام العقارات الخاصة بعلاج المرض مثل الاستاتينس والالتزام بالنظام الغذائى منخفض الدهون مع ممارسة الرياضة.