

## Correlation between Anti cyclic-citrullinated-peptide and rheumatoid Factor Antibodies “levels in” Patients with from Rheumatoid Arthritis

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### Abstract

To identify diagnostic utilities of Anti citrullinated protein (ACCP) and Rheumatoid factor (RF) are autoantibodies (Abs directly against an self-individual antigens), Analytical study. The questioner reported for 50 patients with RA were collected from Department of Rheumatology, AL-Sader Teaching Hospital. Serum levels of RF & (ACCP) Abs were determined by enzyme - linked immunosorbent assay”, while the level of erythrocyte sedimentation rate (ESR) was determined by westergreen method. Distribution of RA occur in females more than males which reached (80%) & (20%) respectively according to the patients group. The patients divided according to the age into three groups (<30), (30-50) & (>50). The percent for these groups were (16%), (52%) & (32%). Among the 50 patients with RA, CCP and ESR 49 patients (98%) tested positive for (ACCP) antibodies, and 22 patients (44%) tested positive for RF. and 37 patients (74%) tested increasing levels of ESR in RA patients. The mean value of (ACCP) & ESR shows highly significance ( $P<0.05$ ), while the RF serum levels increase significantly. Demonstrate the (ACCP) antibodies assay is a useful test for diagnosing RA. However, the use of RF and (ACCP) auto- antibodies in combination further elevated the diagnostic value for RA.

**Keyword:** Rheumatoid arthritis, Anti cyclic- citrullinated peptide, Rheumatoid factor.

### الخلاصة

للتعرف على استخدام الانتي سايكل للبروتين سيتروليناند ببتايد والعامل الروماتزم ونسبة ترسيب كريات الدم الحمراء في تشخيص المرضى المصابين بالروماتزم الرثياني.

جمعت خمسين عينة لمرضى مصابين بالروماتزم الرثياني من قسم الامراض الثانيه في مدينة الصدر الطبية التعليميه , حدوث نسبة مستوى عامل الروماتزم والانتي سايكل سيتروليناند ببتايد بطريقة الامتزاز المناعي المرتبط بالانزيم , بينما تم قياس نسبة ترسيب الخلايا الحمراء بطريقة الويسترجرين.

بينت الدراسه بأن نسبة توزيع المرضى المصابين بالروماتزم في النساء اكثر من الرجال 80%, 20% بالتتابع حسب مجاميع المرضى. قسمت المرضى الى ثلاث مجاميع حسب العمر أقل من (30), (30 – 50) وأكثر من (50) أظهرت نسبة هذه المجاميع كالتالي 16%, 52%, 32% , كما بينت الدراسه بأن نسبة (49) 98% كان لديهم فحص موجب للانتي سايكل ستروليناند ببتايد ونسبة (22) 44% لديهم فحص لعامل الروماتزم و نسبة (37) 74% كان لديهم ارتفاع في مستوى ترسيب كريات الدم الحمراء .

أخيراً أظهرت النتائج بأن متوسط القيم للانتي سايكل ستروليناند ببتايد وترسيب كريات الدم الحمراء كان عالي المنويه ( $P < 0.05$ ) بينما اظهرت النتائج ارتفاع معنوي في مستوى عامل الروماتزم. لذلك فأن استخدام سايكل ستروليناند ببتايد يعتبر فحص مفيد لتشخيص الروماتزم, وكذلك استخدام عامل الروماتزم والانتي سايكل ستروليناند ببتايد ممكن ان يعطي قيمة تشخيصيه للروماتزم الرثياني.

**الكلمات المفتاحيه:** الروماتزم الرثياني, انتي سايكل ستروليناند ببتايد, عامل الروماتزم.

## Introduction

Rheumatoid arthritis (RA) is an inflammatory disorder issue that described by polyarthritis and progressive joint harm “and disability, immunological” anomalies, chronic inflammation. It influences about 1% adult persons worldwide, furthermore it affects one in each thousand of a kids and adolescent. RA usually occurs in females and influences about 2-3 times more than men, also increases flare-up in pregnant females about 70% and increase signs and symptoms after childbirth *Scott, DL(2010)*. The etiology of RA is not referred to, which is categorized as one of the immune system disorders *Cassim, B. (2002)*. It is connected with lessened future and a noteworthy of ceaseless insufficiency and incapacitate, and circumstances turn out to be more serious during the time. Numerous studies have demonstrated that development treatment involves the utilization of right on time ,forceful treatment, that includes antibody against cytokines leading to enhance patient's personal satisfaction, facilitate clinical manifestations, retarded the progression of joint damage ,and deferred disability *Goldbach-Mansky,(2003)*. Anti – cyclic citrullinated peptides antibodies (ACCPAs) are antibodies, play an important role against self-peptides and protein called citrullinated. These antibodies are considered as autoantibodies against own individuals proteins. That available in serum of patients with rheumatoid joint pain. Clinically, cyclic citrullinated peptides (CCP) are frequently used to distinguish these antibodies with high affectability in patient serum or plasma (then referred to as anti–citrullinated peptide antibodies). One of the initially recognized antigens for these antibodies is citrullinated filaggrin *Detrick, Barbara (2006)*. Truly, the another specificity of the same antibodies is described as “anti-keratin antibodies (AKA) and anti-perinuclear factor (APF) *Schellekens, GA (1998)*.

RF is a immunizer coordinated against the Fc part of IgG molecules *Tsavaris N. (2007)* and found in each immunoglobulin subclass (IgM, IgA and IgG) *Elaine Moore (2007)*. The specificity of RF in RA is low (70-80%) *Sharif S. (2007)*, *Suzuki K. (2003)* . The Fc part of IgG molecules testing for the mix of anti-CCP antibodies and IgM RF might be better to exclude the finding of RA than is achievable by testing for either immune response alone *Dubucquoi S. (2004)*.

## Aims of the study

Therefor the aim of this study was to determine the levels of ACCP and RF as biomarkers for diagnosis of rheumatoid arthritis.

## Materials and Methods

### Patients

The first group included of the period June/2016 to March/2017, fifty patients with Rheumatoid joint pain (40 female and 10 male) with ages groups at (21-70) years have been taken from (Al-Sadder Medical City/Najaf ), while the second group of control involved of 20 individuals are have healthy of RA and normal in age and gender with patients, and have no history for any joint damage issues.

### Samples collection and assay procedure

Blood was collected from each individual ,3 – 5 ml of venous. The blood was allowed to clot naturally at room temperature, and then the sera were separated after centrifugation for 3 – 5 min. at (3000 rpm). The sera were frozen at (-20 C°)

until time of examination. Assessments of ELISA kit ACCP ELISA kit (Aeskulisa/ Germany) ,RF ELISA Kit (AeskulisaRF /Germany) detection of rheumatoid factors (RF) in human serum, utilizing industrially accessible and performed as suggested in leaflet with kit. The level of erythrocyte sedimentation rate (ESR) were controlled by westergreen strategy.

### Statistical Analysis

Results were communicated as mean  $\pm$  standard mistake (SE), understudy t-test, ANOVA and Pearson correlation were utilized to analyze results by utilizing SPSS variant 22, p-value  $\leq 0.05$  was considered significant Wayne, w.(2010).

### Results

Table (1) illustrates the distribution of information for (50) patients with rheumatoid joint pain, 10(20%) were male and 40(80%)were female with a range of (21-70) years . The age group divided into three categories <30(16%) , 30-50(52%) , and >50(32%) . Likewise, the patient according severity of the disease as the accompanying : Mild 10(20%) , moderate 12(24%) and sever 28(56%).

**Table(1): Information for Rheumatoid arthritis patient.**

characteristic	Number	Prevalence%
Number of patients	50	100%
male	10	20%
female	40	80%
<b>Age(year)</b>		
range	70 - 21	
30>	8	16%
30 - 50	26	52%
50<	16	32%
<b>Severity of disease</b>		
mild	10	20%
moderate	12	24%
severe	28	56%

A summary of the patients clinical symptomology was collected from a questionnaire presented in table (2) includes join pain , joint swelling , stiffness ,and fever were predominant symptoms, as shown below.

**Table(2): Distribution of Rheumatic patients according to the Clinical Symptoms.**

Clinical symptom	Number	Prevalence %
Joint pain	50	100%
Joint swelling	31	62%
Joint stiffness	25	50%
fever	28	56%

The Anti-CCP , RF and ESR mean value were 701.09 , 21.03 and 33.22 respectively, while in control group the Anti-CCP , RF and ESR , mean value were reached 2.12, 6.54 and 17.09 respectively ,as shown in table (3):

**Table (3): The concentrations A of CCP, RF and ESR in patients and control groups.**

Parameters	Patients Mean $\pm$ std.Deviation No(50)	Control Mean $\pm$ std.Deviation No(20)	F	P .value
ACCP	701.09 $\pm$ 711.31	2.12 $\pm$ 1.51	19.143	0.000
RF	21.03 $\pm$ 36.4	6.54 $\pm$ 17.68	5.760	0.019
ESR	33.22 $\pm$ 14.477	17.09 $\pm$ 11.002	20.115	0.000

The distribution of Anti-CCP, RF, and ESR according to the age, gender ,disease duration and disease stage respectively as shown in tables (4,5,6,7):

**Table (4):Estimation the concentrations of the study Parameters According to age group.**

Parameters	<30 Mean $\pm$ Std. Deviation No(8)	30_50 Mean $\pm$ Std.Deviation No(26)	>50 Mean $\pm$ Std. Deviation NO(16)	P. value
ACCP, IU	689.05 $\pm$ 977.04	634.5 $\pm$ 502.4	808.3 $\pm$ 820.7	0.053
RF, IU	25.65 $\pm$ 62.59	21.41 $\pm$ 31.30	17.56 $\pm$ 21.19	0.429
ESR, mm/hour	26.40 $\pm$ 18.5	33.62 $\pm$ 12.88	36.88 $\pm$ 13.36	0.824

**Table (5): Man value of Serum Concentration for the study Parameter According to gender.**

Parameters	Male Mean $\pm$ Std. Deviation No(10)	Female Mean $\pm$ Std. Deviation No(40)	p· value
ACCP	471.6 $\pm$ 307.4	758.4 $\pm$ 772.6	0.757
RF	2.74 $\pm$ 2.76	25.60 $\pm$ 39.44	0.862
ESR	34.10 $\pm$ 11.06	33.00 $\pm$ 15.32	0.198

**Table (6): Mean value of serum concentration for the study Parameter According to disease duration.**

Parameters	1-5(year) Mean $\pm$ std.Deviation No(41)	>5(year) Mean $\pm$ std.Deviation No(9)	p· value
ACCP	598.09 $\pm$ 646.23	1066.25 $\pm$ 839.18	0.353
RF	18.84 $\pm$ 37.86	28.79 $\pm$ 30.98	0.271
ESR	32.97 $\pm$ 15.04	34.09 $\pm$ 12.84	0.800

**Table(7):Mean value of Serum Concentration for the study Parameter According to disease stage.**

Parameters	Mild Mean $\pm$ Std. Deviation No(10)	Moderate Mean $\pm$ Std.Deviation No(12)	Severe Mean $\pm$ Std.Deviation No(28)	p· value
ACCP	575.5 $\pm$ 405.2	945.8 $\pm$ 955.4	629.7 $\pm$ 658.8	0.258
RF	23.69 $\pm$ 37.24	33.69 $\pm$ 54.86	13.94 $\pm$ 22.20	0.075
ESR	31.30 $\pm$ 13.06	32.08 $\pm$ 13.33	34.48 $\pm$ 15.810	0.832

The level of Anti CC-P was correlated with RF and ESR with p.value(0.000 , 0.001) respectively, also was RF correlate with ESR in P.value (0.032) as shown in table (8).

**Table (8): correlation among study parameter.**

parameters	R	P• value
ACCP VS RF	0.589 <sup>+</sup>	0.000
ACCP VS ESR	0.464 <sup>+</sup>	0.001
ESR VS RF	0.304 <sup>+</sup>	0.032

**Table(9): Determination of rheumatic patient according to positive and negative result in A-CCP and RF.**

RF	A-CCP	Number	Percent%
Positive	Positive	21	42%
Positive	Negative	1	2%
Negative	Positive	28	56%

## Discussion

In the present study , the Anti – CCP is significance as , analytic and prognostic marker for RA has been illustrated broadly in the developed world . Even more so late ,A-CCP antibodies have been distinguished in the of RA patients . These antibodies have shown high diagnostic specificity and generally increase the level sensitivity *Vallbracht, I. (2004)*. Ordinarily , the serology test routinely utilized as a part of RA diagnose which includes serum RF levels. However , it has minimal predictive quality in the inclusive community ,since the general disease prevalence is moderately low. The high level of particular self-antibodies for the conclusion of RA ,A-CCP antibodies ,were found in *Nienhuis RLF, Mandema E. (1964)*. Joint agony, swelling and solidness were the predominant symptoms. the disease essentially influenced the little joints of the hands, however every one of the joints in the body were influenced to some extent *Aysha, h. k. (2012)*. In our study, the positive results for rheumatoid factors and (ACCP), that affect sickness prognosis and are considered as autonomous indicators for erosive advancement ,and the relation of that antibodies increase erosive damage.

Many studies indicate a positive RF levels and considered vital indicator for join damage throughout development of disease during years. *Jansen et al,(2001)* presumed the progression of radiographic at one year was anticipated with positive result of RF. Like most studies, we recommended the prognostic estimation of RF levels. It creates the impression the (ACCP) antibodies level is considered very important like RF levels. *Vencovsky et al,(2003)* introduced that (ACCP) consider better predicting than RF at Larsen score in about two years. Imminent investigation for early RA followed up for a

long time the (ACCP) immune response resulted in association with RF ,however being considered more benefit than RF as indicator for high risk disease. This study demonstrated nearness of RF and (ACCP) antibodies was connected with high probability of erosion sickness. The joined utilization of (ACCP) and RF was more noteworthy in it is specificity as indicator for erosive damage than RF or (ACCP) alone.

## Conclusion

The ACCP antibodies was excellent immunological markers for RA, and appropriate as a direct approach diagnostic test for RA. In RF and ACCP seronegative patients, may be useful in affirming the finding of RA. The combination of RF and (ACCP) tests instead of RF or (ACCP) to get the best results in RA determination.

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