Original Article

Comparison of sensitivity and specificity of anti-CCP and anti-MCV antibodies in an Iranian cohort of patients with rheumatoid arthritis

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Abstract

Background: Anti-CCP is a test commonly used for the diagnosis of rheumatoid arthritis. The aim of this study was to determine the diagnostic values of ACCP compared to anti-MCV in rheumatoid arthritis patients in north of Iran.

Methods: The serum samples of 150 RA patients and 75 controls, with the mean age of 49.6±11.8 and 48.8±12 years respectively, were tested using the commercially available ELISA kits for ACCP and anti-MCV. Sensitivity, Specificity were determined and Roc curve were used for comparison between these two groups.

Results: The sensitivity of ACCP versus anti-MCV was 85% and 81%, respectively. Specificity was 96% and 95%, respectively. In the RA patients, ACCP was positive in 127 (84.7%) and anti MCV in 121 (80.7%) cases. In the control group, these parameters were positive in 3 (4%) and 4 (5.3%) ($p<0.0001$ and $p<0.0001$, respectively). The correlation coefficient for ACCP and anti-MCV was calculated at 0.63 ($p<0.001$). The area under the curve for ACCP was 0.941±0.015 ($p<0.001$), anti-MCV was 0.902±0.02 ($p<0.001$). The measure of agreement (Kappa) for these variables was 0.81. In these patients, there was no correlation between DAS28 and the positivity of these tests.

Conclusion: It was concluded, compared to ACCP, anti-MCV has approximately the same accuracy for the diagnosis of rheumatoid arthritis and it does not have additional value.

Keywords: Rheumatoid arthritis, ACCP, Anti-MCV

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Rheumatoid arthritis (RA) is the most common chronic, progressive inflammatory disorder affecting synovial joints and leading to inflammation-induced comorbidities. The global prevalence of RA ranges between 0.5-1%, mostly in young women and elderly people (1). The currently laboratory diagnostics of RA particularly early RA, is based on a highly specific marker of the disease such as antibodies against citrullinated proteins. The positive test for anti-cyclic citrullinated protein (ACCP) antibody is now used as a classification criterion of RA (2). ACCP test may help predict the transformation of undifferentiated arthritis into RA. The probability of developing RA from undifferentiated arthritis in patients with positive ACCP is 90%, whereas 30% in those with negative test (3). In a study, the positive predictive value for progression to RA was 80% and this was increased when 2 or 3 other tests were used together (4).

RA patients are now divided into two groups those with positive and negative ACCP antibodies. ACCP positivity predisposes individuals to more advanced course of the disease, with extensive bony erosions, accelerated atherothrombotic disease and worse overall prognosis (3). From the different types of ACCP antibodies, ACCP2 is found to be the most sensitive and specific diagnostic marker (3-5). Most citrullinated proteins (e.g. fibrinogen, histones) are associated with this antibody (3).

Anti-mutated citrullinated vimentin (Anti-MCV) is another anti-citrullinated antibody reacting with mutated citrullinated vimentin. In fact, it is the Sa antigen and is expressed in fibroblast like synoviocytes (6). Also citrullinated fibrinogen is another antigen that is detected in synovial tissue of rheumatoid arthritis inflamed joints (4). There are many studies in comparing anti-MCV and ACCP for their diagnostic value in rheumatoid arthritis. In addition to these tests, also anti-MCV especially correlate with higher levels of DAS28 and joint damage (7-10, 12). The purpose of this study was to compare the two tests (ACCP2 and anti-MCV) in an Iranian cohort of patients with RA.

Methods

This cross-sectional study was performed from January to June, at the Rheumatology Clinic, Babol, Iran. We enrolled 150 patients with RA, who met the ACR 1987 classification criteria. The duration of RA ranged from 6 months to 6 years. All patients were treated with prednisolone, hydroxychloroquine and methotrexate. None of the patients received therapy with biologic agents. As controls, we recruited 75 subjects including those patients with low back pain, osteoarthritis, gout, and individuals with non-RA rheumatic diseases.

From the total 225 subjects, 8 ml blood samples were collected and processed at the laboratory according to specifications. After centrifugation, aliquots were separated and frozen. ACCP, and anti-MCV tests were performed on all samples. The ACCP2 test was done by using Euroimmne Kit, Lubeck, Germany. The level of greater than 5 RU/ml was considered positive. The anti-MCV test was done by using Orgentic Diagnostika kit, Hamburg, Germany. The level greater than 20 IU/ml was considered positive. The positivity levels were according to manufacturer's instructions. In all 150 RA patients, DAS28 were calculated. DAS28 ≤2.6 consider inactive disease, 2.6-3.2 as mild, 3.2-5.1 moderate and more than 5.1, high disease activity. The data were collected and analyzed using SPSS version 18. The quantitative variables were analyzed with student's t-test, and qualitative variables with chi-square test. ROC curve was used for determining the sensitivity and specificity of the laboratory markers. The area under the curve of each test was calculated and compared with each other. Correlation coefficient and measure of agreement (Kappa) was determined for both tests. Significance was assumed at $p<0.05$.

Results

From the 150 patients with RA, 116 were females (77.3%) and 34 males (22.7%). The mean age was 49.6±11.8 years. The control group consisted of 17 males (22.7%) and 58 females (77.3%) with the mean age of 48.8±12 years. In the RA patients group, ACCP was positive in 127 cases (84.7%) and anti MCV in 121 cases (80.7%). In the control group, these parameters were positive in 3 (4%) and 4 (5.3%). In the RA group distribution of positive cases according to DAS28 is presented in table 1. Thirty-three patients (22%) were in remission, 21 (14%) had mild disease activity, 69 (46%) - moderate activity and 27 (18%), high activity.

There was no statistically significant difference between groups. Table 2 depicts sensitivity, specificity, positive and negative likelihood ratio of ACCP, anti-MCV. In RA patients group, both ACCP and anti MCV were negative in19 cases, both were positive in 117 cases, positive ACCP and negative anti-MCV in 10 cases and negative ACCP and positive anti-MCV in 4 cases. The Kappa measure of agreement was 0.810 for these results. The correlation coefficient for ACCP and anti-MCV was calculated at 0.63 ($p<0.001$).

Table 1. Distribution of positivity and negativity of ACCP, Anti-MCV according to DAS28

Test		DAS 28								P-value
		Remission		Low activity		Moderate activity		High activity		
		N	%	N	%	N	%	N	%	
Anti-MCV	Positive	25	20.7	17	14	56	46.3	23	19	0.832
	Negative	8	27.6	4	13.8	13	44.8	4	13.8	
ACCP	Positive	23	18.1	63	49.6	15	11.8	26	20.5	0.109
	Negative	4	17.4	6	26.1	6	26.1	7	30.4	

Table 2. Diagnostic performance of ACCP and Anti-MCV

	Sensitivity	CI	Specificity	CI	PPV	CI	NPV	CI	LR+	CI	LR-	CI
ACCP	85%	79-90	96	92-100	98	95-100	76	67-84	21.17	6.97-64.28	0.16	0.11-0.23
Anti-MCV	81%	74-87	95	90-100	97	94-100	71	62-80	15.12	5.81-39.37	0.2	0.15-0.28

The ROC curves for ACCP, anti MCV were drawn and area under the curve for these tests was measured. The area under the curve for ACCP was 0.941±0.015 (p<0.001), anti-MCV was 0.902±0.02 (p<0.001), which means that both tests were instrumental for RA diagnosis (figures 1, 2).

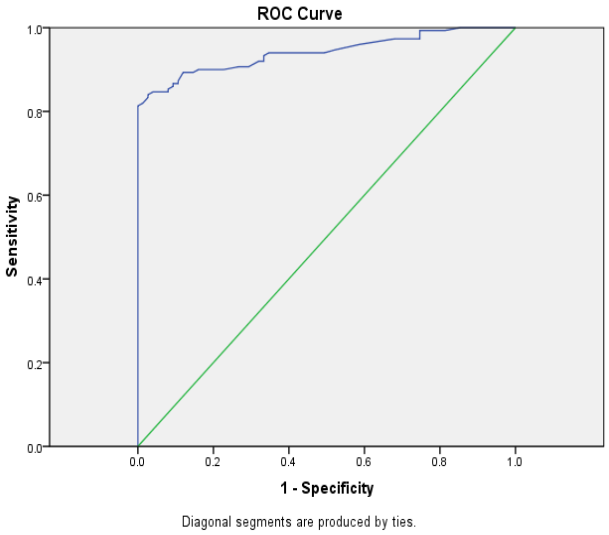

Fig.1 ROC curve of ACCP in RA patients

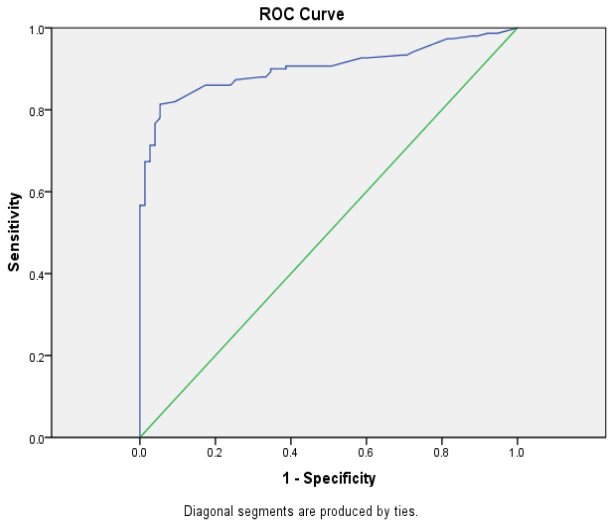

Fig.2 ROC curve for Anti-MCV in RA patients

Discussion

In this study, we compared the diagnostic values of ACCP and anti-MCV. The aim of this study was to distinguish more informative test for diagnosing RA. Previous studies yielded controversial results without a definite agreement to conclude which test is more accurate. There was also no large study in an Iranian cohort of RA patients. In a study by Van der Linden et al. for predicting from undifferentiated arthritis to RA, the ACCP test had very good specificity and PPV. Anti-MCV did not seem to be more informative, and adding RF and anti-MCV tests to ACCP2 did not enhance the diagnostic value of the laboratory test and it concluded that single test was enough (13).

High specificity, more than 90% PPV for ACCP and a poor outcome for ACCP positive RA patients was reported in a review published in 2010 (3). In a recent meta-analysis, 16 studies on anti-MCV were analyzed; sensitivity, specificity, positive LR, negative LR and diagnostic odds ratios (ORs) were estimated to be 0.77, 0.89, 7.24, 0.28 and 29.66 respectively (14).

In another study, sensitivity of anti-MCV was reported to be 79.6% and specificity 96.6%; test positivity was accompanied with a higher DAS28 (9). This was also confirmed in a study by Syversen SW et al. who reported more advanced joint damage in those with anti-MCV positivity (12).

In most of the published works that we studied, the sensitivity of anti-MCV was somehow higher than ACCP but ACCP was more specific (4, 6-8, 11). The same results have been mentioned in some other studies that Ernest Wagner et al. referred to. They found that in RF negative patients, the sensitivity of anti-MCV is higher (43.8% versus 30%) (8).

Our study did not show the significant differences between sensitivity and specificity of ACCP and anti-MCV (sensitivity 85%, 81%, specificity 96% and 95%, respectively). In 117 of our cases (78%) ACCP and anti-

MCV were positive, and in 19 patients (12.7%) both were negative. The analysis of the above results yielded kappa of high agreement between these two tests (kappa=0.81), and correlation coefficient of 0.63 (p=0.001) which means that both tests have similar value. In other words, ACCP and anti-MCV positivity usually coincide.

In Sghiri et al.'s study, there was also a significant correlation between anti-MCV and ACCP (6). However, in our study, in small number of cases, this was not true: [10 cases (6.7%) had positive ACCP and negative anti-MCV, while 4 cases (2.7%) had negative ACCP and positive anti-MCV]. It was slightly different in the Nicase-Roland et al.'s study in which the number of positive anti-MCV in ACCP cases was equal to the number of positive ACCP in anti-MCV negative patients (15).

In the ROC analysis, the level for each test with 100% specificity was determined. This was 9.8 Ru/ml for ACCP (2 times of the laboratory cut-off point) and 89.5 u/ml for anti-MCV (4 times of the laboratory cut-off point). The sensitivity of the tests was 81% and 57%, respectively. The latter means that ACCP with the level of 2 times more than normal and 81% sensitivity is specific for diagnosis of RA. But, for anti-MCV, this level is 4 times more than normal with a sensitivity of 57%. Below these levels, anti-MCV has less specificity. This might be a reason that anti-MCV has been introduced as a new biomarker for diagnosis of ankylosing spondylitis (16). Positive anti-MCV was also reported in SLE, Sjogren syndrome, psoriatic arthritis, EBV and HCV infected patients (8, 11). Because of low number of non- RA controls in our study, we obtained these results.

In conclusion, our study suggests that ACCP is an informative diagnostic test for RA. anti-MCV does not have additional value. This statement is based on the somehow more sensitivity and specificity and the results of kappa, indicating positivity of ACCP in patients positive for anti-MCV and *vice versa*.

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Conflict of Interest: None declared.

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