Comparison of the anti-cyclic citrullinated peptide and rheumatoid factor in rheumatoid arthritis at an arthritis center

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Rheumatoid arthritis (RA) is a systemic inflammatory disease that frequently starts at the peak of productive life and causes irreversible joint damage, disability, and premature mortality. To improve the result it is important to diagnose and treat the disease early.[1] The diagnosis of RA is usually based on the ACR (American College Of Rheumatology) classification criteria that include clinical manifestations, and the presence of the IgM rheumatoid factor (IgM-RF). Rheumatoid factor is detected in 50-80% of the patients. Unfortunately, it is not specific (80-90%) since it can be detected in other autoimmune diseases, in chronic infections, and in 5-10% of healthy individuals. In recent years, several novel autoantibodies have been described in RA. In particular, antibodies against citrullinated proteins, such as filaggrin and a commercial synthetic circular peptide (cyclic citrullinated peptide or CCP). The presence of antibodies to CCP provides important diagnostic information in patients with RA.[2] The objective of this study was to evaluate the clinical utility of anti-CCP in RA by determining the sensitivity, and specificity of anti-CCP antibodies prospectively in RA patients using a control group of patients with other connective tissue diseases, and inflammatory arthritis. We also determined the correlation between the anti-CCP antibodies level, and levels of IgM-RF and total serum IgG.

One hundred and forty-six adult consecutive patients attending the out patient clinic at the Arthritis Centre, Riverside Campus of the Ottawa Hospital in Ottawa, Canada were studied from October 2005 to January 2006. Patients were divided into 2 groups according to ACR classification criteria, RA (n=97) and non-RA patients (n=48). The non-RA patients included psoriatic arthritis (n=20), systemic lupus erythematosus (SLE) (n=19), juvenile rheumatoid arthritis (n=3), seronegative spondyloarthritis (n=2), polymyalgia rheumatica (n=2), Sjogren's syndrome (n=1), relapsing polychondritis (n=1), and palindromic rheumatism (n=1). The Ottawa Hospital-Research Ethics Board approved the study. The IgM-RF, total IgG, and anti-CCP were measured in patients' serum samples. The IgM-RF was measured by nephelometry (Immage®, Immunoassay System, Beckman-Coulter Inc., California, USA) (normal upper limit: 20 IU/L). On the same instrument, IgG was measured with a lower detection limit of 0.33 g/L and effectively no upper limit. Anti-CCP of IgG isotype was measured by ELISA immunoassay (Euro Immune®, second generation, Mainz, Germany). This method has a lower limit of detection of 1 RU/mL and upper limit of 200 RU/mL. All methods were used according to manufacturer's instructions. Sensitivity, and specificity in RA versus non-RA were calculated using Medmath, and SPSS Software (version 10) with 95% confidence interval for the sensitivity and specificity. The correlation between quantitative anti-CCP with quantitative IgM-RF, and IgG immunoglobulin was determined via a correlation Pearson's product-moment coefficient. Any results beyond the measurement limits of the tests were excluded in order to carry out the linear regression analysis.

Sixty-three out of 146 patients tested positive for anti-CCP antibody at more than 5 units reactivity, 58/63 had RA. This translated into a sensitivity and specificity of anti-CCP reactivity for the diagnosis of RA of 51% (95% CI 40.8-61.1), and 98% (95% CI 88.7-99.6). This compared with the sensitivity and specificity of IgM-RF in RA of 47% (95% CI 38.9-60.2), and 89% (95% CI 76-96.4). In the RA patients, 42/97 (43%) were both anti-CCP, and RF positive, and 15/97 (16%) were anti-CCP positive, and RF negative. This represents the percent of the patients that could be picked up by a positive anti-CCP test alone (**Table 1**).

Rheumatoid arthritis and other autoimmune diseases are characterized by a state of immune hyperactivity. We examined whether the anti-CCP antibodies were associated with the hyperactivity. We investigated the linkage between IgM-RF, and anti-CCP by correlation coefficient and found a significant positive correlation between both tests (R=0.37, p<0.001), keeping in mind the limited number of patients remaining after the elimination process, documented in an earlier reporf.[3] Anti-CCP antibodies are in the IgG class. We investigated the linkage between anti-CCP, and IgG by correlation. In RA patients, there was a positive correlation between

Table 1 - Percent of the anti-CCP and RF in rheumatoid arthritis patients.

Test	Anti-CCP	
	Positive (n=34) n (%)	**Negative (n=63)** n (%)
Rheumatoid factor		
Positive, (n=51)	42 (43.3)	6 (6.2)
Negative, (n=46)	15 (16.5)	34 (35.1)

Anti-CCP - anti cyclic citrullinated peptide, RF - rheumatoid factor

anti-CCP, and IgG (R=0.3, $p<0.003$. One (5%) of the 18 patients with SLE had positive anti-CCP antibodies. This is lower than the percentage that has been detected in previous studies of 10-15%.[4] Among 20 patients with psoriatic arthritis, 3 (15%) had a positive anti-CCP, and one (5%) had a positive IgM-RF, which are the same rates reported in other studies. Anti-CCP is an assay with a high specificity for the diagnosis of RA; 99% using healthy individuals as a controlled group. Over the past years many studies have been reported, evaluating the specificity of anti-CCP using controls of both categories: healthy individuals, and patients with other autoimmune diseases. They reported specificity ranging from 96-99%.[5] Our study found a similar specificity of 98% with the autoimmune diseases control group. Five percent of SLE patients, and 15% of psoriatic arthritis patients had a positive test, which is similar to previous reports. Rheumatoid factor, and other autoimmune diseases are characterized by a state of immune hyperactivity. We examined whether the anti-CCP antibodies were part of the hyperactivity, and found a correlation between the level of anti-CCP response, and the total IgG. Our study has limitations; we did not explore the relationship between anti-CCP antibodies and measures of disease severity such as the presence of joint space narrowing and erosions. We did not study a large group of normal controls to refine our estimates of sensitivity and specificity. At the Ottawa Hospital, the cost for the IgM RF is $4.30 Canadian/test and anti-CCP is $8.80 per test.

In conclusion, despite the more specific association with RA, anti-CCP antibody does not carry any significant advantage over RF in patients who are RF positive. However, the test might be useful in patients who are IgM-RF negative, but have features clinically suggestive of rheumatoid arthritis. Further study is needed to confirm the correlation between the anti-CCP level with the IgM-RF in Saudi population.

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