Anti-citrullinated peptide antibody: death of the rheumatoid factor?

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ABSTRACT

- Early diagnosis and treatment of rheumatoid arthritis (RA) is necessary to prevent joint damage and long-term disability.
- High rates of false-negative and false-positive results of the rheumatoid factor (RF) test make it generally unhelpful in the early diagnosis of RA.
- A new clinical test for RA — the anti-citrullinated peptide antibody (ACPA) test — is now widely available in Australia. Owing to its high specificity (95%), a positive ACPA test result usually confirms a diagnosis of RA in a patient with undifferentiated inflammatory arthritis.
- The superior specificity of the ACPA test provides an argument for it to replace the RF test in the primary care setting. Performing both tests adds little to the use of the ACPA test alone.
- An early diagnostic opinion from a rheumatologist is still recommended, as the ACPA and RF tests frequently return negative results in early RA.

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Rheumatoid arthritis (RA) is the most common cause of inflammatory arthritis, affecting around 1% of the general population. Untreated, it causes irreversible joint damage and disability in most patients, many of whom are young adults. Clinical features of early RA include symmetrical small joint pain, swelling and morning stiffness, lasting for more than 6 weeks.[1] Early treatment with disease-modifying antirheumatic drugs (DMARDs) such as methotrexate prevents joint damage and disability.[2] This recently accepted treatment paradigm has placed greater importance on accurate early diagnosis of RA. Misdiagnosis carries risks not only of undertreatment, but also of exposing patients to unnecessary toxicity. The diagnostic pitfalls of the rheumatoid factor (RF) test are well described,[1] and a presumptive diagnosis of early RA has been largely based on history, and symmetrical synovitis of small joints on examination. A new assay — the anti-citrullinated peptide antibody (ACPA) test — is now available. Recently reported clinical studies in various populations confirm that the ACPA test has a high specificity. We describe its potential role in the early diagnosis of RA.

The problem with RF

The traditional autoantibody test of choice in RA is the RF test, which has been in clinical use for more than 50 years. This test detects the presence of RF — autoantibodies directed against the constant region (Fc) of the human IgG antibody. RF is included in the current classification criteria for RA.[3] A large body of evidence associates positive RF status with various disease outcomes in RA — poorer prognosis, progressive radiological damage, and extra-articular manifestations.[1] Nevertheless, the diagnostic utility of RF is compromised by its poor specificity. A false-positive test result may occur in patients with various infections (including hepatitis C), other non-RA autoimmune conditions, and malignancies.[1] RF also occurs in about 5% of healthy people, and increases in prevalence with age.[4]

What is the ACPA test?

ACPAs are quite different autoantibodies to RF. The ACPA test, also known as the anti-cyclic citrullinated peptide (anti-CCP) antibody test, is an enzyme-linked immunosorbent assay that tests for the presence of antibodies that recognise specific antigens containing citrulline.[5,6] Citrulline is a non-standard amino acid that is created by enzymatic modification of arginine (a process known as citrullination). The current version of the anti-CCP test has been commercially available since 2000.

Performance characteristics of ACPA and RF tests

There are numerous reports of the characteristics of the ACPA test in cohorts with established RA and early arthritis, as well as in healthy individuals. A recent systematic review[7] and a separate meta-analysis[8] concluded that the specificity of the ACPA test for RA was 95% (95% CI, 94%–97%), compared with 85% specificity of the RF test for RA. The sensitivities of the two tests for RA were similar — 67% for ACPA and 69% for RF. Similar results were found in an Australian series testing ACPA as a quality assurance exercise in parallel with RF testing in 248 patients, of whom 43 had RA. The sensitivity of the RF test was 74%, and the specificity 77%. With ACPA testing, sensitivity was 81%, and specificity 95% (A D S, unpublished data).

Clinical utility of ACPA and RF tests

A working knowledge of a test's statistical characteristics is necessary for its rational use in clinical practice. The usefulness of a diagnostic test is efficiently measured by its likelihood ratio (LR), which brings together the information from sensitivity and specificity into a single figure. Understanding LRs is the key to understanding the utility of ACPA and RF tests.

The LR measures the ability of a test to alter the likelihood of a diagnosis. When a test with a large positive LR (LR+) returns a positive result, the post-test probability of the diagnosis will be significantly higher than the pre-test probability — in other words, the diagnosis will be much more likely. When a test has a small LR+ and returns a positive result, the difference between pre-test and post-test probabilities will be small — in other words, the test adds little to the pre-test assessment. The most useful diagnostic tests have an LR+ that is greater than 10, meaning that the post-test odds of diagnosing disease are more than 10 times the pre-test odds when the test returns a positive result. In terms of probability, this corresponds to an approximately 45% absolute increase in the probability of a diagnosis.[9]

A negative test result may be useful in reducing the chance of diagnosing a disease. The usefulness of such tests is measured by the negative LR (LR–), with the most useful tests having an LR– that is less than 0.1. These tests reduce the pre-test odds of diagnosing disease by more than 10-fold. More detailed explanations of LR and its uses are available elsewhere.[10]

Post-test probabilities of a diagnosis of RA when positive and negative RF and ACPA test results are obtained for a range of pre-test probabilities

	Positive test result		Negative test result	
Pre-test probability	RF	ACPA	RF	ACPA
1%	4.7%	11.2%	0.4%	0.4%
10%	35.1%	58.1%	4.1%	3.8%
25%	61.8%	80.6%	11.2%	10.7%
50%	82.9%	92.6%	27.5%	26.5%
75%	93.6%	97.4%	53.3%	51.9%
90%	97.8%	99.1%	77.4%	76.4%

RA = rheumatoid arthritis. RF = rheumatoid factor. ACPA = anti-citrullinated peptide antibody. ◆

The meta-analysis of the ACPA testing in RA showed that its LR+ was 12.46 (95% CI, 9.72–15.98) and LR– was 0.36 (95% CI, 0.31–0.42).[8] This suggests that it is a useful test when it returns a positive result. In contrast, the LR+ for the RF test in RA was 4.86 and the LR– was 0.38. This suggests that the RF test is sometimes diagnostically useful when it returns a positive result. Based on their respective LR– values, neither the ACPA test nor the RF test are useful in RA when they return a negative result (as both have an LR– that is greater than 0.1).

Clinical examples

The following examples demonstrate the variable utility of RF and ACPA tests in different clinical situations (see Box). They highlight the importance of determining the pre-test probability of a diagnosis (estimated from the clinical assessment) and how it is affected by the test's LR. The post-test probability is dependent on these two factors. The treatment decision threshold depends on several factors, and these examples do not take into account the complexities of differential diagnosis.

- Population screening tests require extremely high sensitivity and specificity to be useful. In this situation, the pre-test probability approximates the community prevalence of RA as being 1%, and neither ACPA nor RF tests are useful.
- In patients with a low pre-test probability of RA (eg, 10%), neither a positive RF test result nor a positive ACPA test result affect clinical probabilities sufficiently to secure a diagnosis. It is worth remembering that a positive diagnosis of RA will usually entail the commencement of DMARD treatment, so there should be reasonable certainty.
- In patients with a moderate pre-test probability of 25%–75%, the effect of a positive ACPA test result is more marked than that of a positive RF test result. The superior usefulness of the ACPA test is also heightened because tests are most needed in this range, where there is genuine clinical uncertainty. Put in a different way, if the confidence to begin DMARD treatment requires an 85% certainty of diagnosis, a positive ACPA test result achieves this when the pre-test probability is 31%, whereas a positive RF test result requires a pre-test probability of 53%. In the clinical context, this difference represents a sizeable group of patients who could begin DMARD treatment earlier or with more confidence. Given the value of starting DMARDs early in RA to prevent joint damage and disability, this is an important difference between the tests.
- In patients with a high pre-test probability of greater than 75% (ie, patients in whom there is minor doubt about the clinical diagnosis), either a positive RF test result or a positive ACPA test result confirms the diagnosis. Negative results at this range lower the certainty somewhat, but should not deter clinicians from a diagnosis of RA, which still remains likely. Furthermore, negative results from either test are generally unhelpful across the range of probabilities. This reflects the poor sensitivities of both tests, which fail to identify the so-called sero-negative RA patient group.

Combining ACPA with RF

The utility of combining ACPA and RF tests has been addressed in few studies. When both tests return positive results, the LR+ is 15.72 and the LR– is 0.46. Thus the test performance of the combination is little better than the ACPA test alone, and adding RF improves specificity at the expense of sensitivity. A positive result from either test (ie, one of the two tests positive or both tests positive) improves sensitivity but reduces specificity, giving an LR+ of 4.32 and an LR– of 0.32. Some patients with RA do have either one or other of the autoantibodies, leading to a suggestion that testing be performed consecutively, starting with RF and progressing to ACPA if the RF test result is negative.[11] The established test characteristics described above, however, do not support such a recommendation. Patients with a positive RF test result and a negative ACPA test result may have a false-positive RF, as this test has a lower specificity.

Cost of ACPA and RF tests

The laboratory cost of an RF test is $11.50 (Medicare Benefits Schedule) and a single ACPA test costs $15.00. Although ACPA testing is more expensive, there may be substantial costs associated with false-positive RF results. These results often entail review by a rheumatologist, including time off work and unnecessary distress for the patient. To our knowledge, a formal cost-effectiveness analysis has not yet been reported.

Conclusions

ACPA testing represents an advance in the assessment of inflammatory arthritis. Its superior specificity and LR+, when compared with RF, provide arguments for its use in the diagnostic work-up of patients with symptoms and signs that suggest an inflammatory arthritis, particularly when there is genuine clinical uncertainty about the diagnosis. RF is a familiar test and has been used in most prognostic studies of RA; however, the RF test does not appear to provide much additional diagnostic information to the ACPA test. We believe that the ACPA test has become the diagnostic test of choice in RA, and that it should replace the RF test in this context. Nevertheless, an early diagnostic opinion from a rheumatologist should still be sought as negative results are frequently obtained from ACPA and RF tests in early RA.

Competing interests

None identified.

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