Rheumatoid Arthritis
Laboratory Markers for Diagnosis and Prognosis

CLINICAL BACKGROUND

Rheumatoid arthritis (RA) is an autoimmune disease that affects mainly the synovial membranes and articular structures and is characterized by chronic, systemic inflammation involving multiple joints. Small joints in the hands and feet are usually affected first, followed by the larger joints. Patients with RA have periodic flare-ups that can lead to irreversible joint destruction. Systemic effects may include damage to organs such as the heart and lungs.

About 1% of the US population has RA, with prevalence being 2 to 3 times higher in women than men. Although the cause of RA remains unknown, the increased risk in family members of patients with RA suggests a genetic component. Environmental or hormonal factors may be involved in perpetuating the inflammatory process and joint destruction.

Treatment with disease-modifying anti-rheumatic drugs (DMARDs) can often ameliorate the disease. Because bone destruction can occur early, with many RA patients showing radiographic evidence of bone destruction in the first 2 years of disease,[1] therapy should be initiated promptly to minimize irreversible joint damage. American College of Rheumatology (ACR) criteria for classification of RA require that 4 of the following 7 conditions be present[2]:

- Morning stiffness in and around joints lasting ≥1 hour (present ≥6 weeks)
- Physician-documented arthritis involving ≥3 joint areas simultaneously (present ≥6 weeks)
- Arthritis of the proximal interphalangeal, metacarpophalangeal, or wrist joints (present ≥6 weeks)
- Symmetric involvement of joint areas (present ≥6 weeks)
- Rheumatoid nodules
- Positive serum rheumatoid factor (RF)
- Radiographic evidence of erosions or periarticular osteopenia in hand or wrist joints

Although these criteria demonstrate good sensitivity (91%-94%) and specificity (89%) in differentiating patients with established RA from control subjects, they are less robust for classifying patients with early inflammatory polyarthritis who may have mild symptoms and signs.[3]

The recently developed anti-cyclic citrullinated peptide (anti-CCP) assay, in combination with RF, may help establish a diagnosis in the early stages of RA when treatment is most effective in preserving function.

This *Clinical Focus* discusses laboratory tests available to assist physicians in differentiating between RA and other conditions that present with polyarticular arthritis, and in monitoring the disease course.

INDIVIDUALS SUITABLE FOR TESTING

- Individuals with symptoms of arthritis not attributed to other conditions
- Individuals with established RA

TEST AVAILABILITY

Table 1 provides a listing of tests that may be useful in the diagnosis, assessment of prognosis, and follow-up of RA.

TEST SELECTION

Because of the low overall prevalence of RA, these assays are not suitable for routine screening of asymptomatic individuals.

Diagnosis

Diagnosis of RA relies primarily on patient history and radiographic evidence of joint damage. Laboratory testing can help differentiate RA from other conditions that manifest with polyarthritis and can be especially useful early in the disease course for establishing diagnosis and prognosis.

RF is the most widely used laboratory marker of RA and is the only one included in the current (1988) ACR classification criteria.[2] RF titer is most often assessed with latex fixation/immunoturbidimetry, which primarily detects IgM RF. IgM RF, as well as IgA and IgG RF, can also be measured individually with specific immunoassays.

Anti-CCP testing is also useful for determining the prognosis of RA, being predictive of disease progression at 3 to 10 years after disease onset. In most studies, anti-CCP positivity at baseline correlates with poor prognosis in terms of radiographic and functional outcome.[20,21] In a study of patients with early arthritis, baseline values of anti-CCP (67%) and RF (69%) showed similar sensitivity for prediction of radiographic progression at 5 years, but anti-CCP showed greater specificity (56% vs 24%).[20] Combining RF with anti-CCP results appeared to help in prediction: RF+/anti-CCP+ patients had greater progression than RF+/anti-CCP– and RF–/CCP– patients at 5 years.

Monitoring Disease Course

CRP and ESR are both acute-phase markers of inflammation sometimes used to monitor RA disease activity. CRP is produced by the liver in response to tissue injury, infection, and inflammation. Levels increase during periods of heightened RA disease activity, but elevations may also reflect inflammation due to other causes, such as infection or injury. The ESR typically rises 24 to 48 hours after an inflammatory stimulus and returns to normal levels gradually thereafter. ESR measurement may help assess disease activity when other clinical and laboratory studies yield equivocal results.[22]

Supportive Testing

A complete blood count with white blood cell differential can help document the mild anemia, leukocytosis, and other hematologic abnormalities sometimes associated with RA. More severe anemia may reflect gastrointestinal bleeding resulting from steroidal and non-steroidal anti-inflammatory drugs. Urinalysis typically yields normal results. Liver and kidney function should be assessed before starting therapy with DMARDs, to establish baseline values, and at intervals thereafter.

TEST INTERPRETATION

The result of each assay should be evaluated in conjunction with clinical and radiographic findings and other serological test results.

RF

Positive RF results are suggestive of RA, but the low specificity precludes a definitive diagnosis. Positive results are also common in patients with other rheumatic diseases and conditions that can mimic RA (Table 3). Negative results are consistent with conditions other than RA but do not rule out RA. Because RF-negative patients may seroconvert, follow-up testing at intervals during the first year of disease may be useful if the initial result is negative. IgA and IgG are both highly specific for RA and are associated with greater disease severity and likelihood of progression. Because of the relatively low sensitivity of IgA and IgG RF, negative results do not indicate absence of RA.

Anti-CCP

Positive anti-CCP results are highly specific for RA and are associated with an aggressive disease course. Although this assay is generally more specific than RF, patients with other rheumatic diseases may rarely have elevated titers (Table 3). Negative results do not rule out a diagnosis of RA but suggest that other rheumatic diseases may be responsible for the patient's symptoms.

Table 3. Reactivity of Rheumatoid Factor (RF) and Anti-Cyclic Citrullinated Antibody (Anti-CCP) Assays in Various Disorders

Population	Sample Size, n	Percent Positive*	
		RF	Anti-CCP (≥20 Units)
RA[5]	231	70	74
Healthy individuals/ blood donors[8†]	154	7	1
SLE[25]	201	13	6
Scleroderma[26]	86	NA	12
Primary Sjögren's syndrome[27†]	134	59	8
Juvenile RA[28]			
Polyarticular onset	77	18	13
Pauciarticular onset	139	7	2
Polymyalgia rheumatica[29]	49	7	0
Mixed connective tissue disease/vasculitis[8†]	103	43	7
Psoriatic arthritis[30]	160	11	7
Non-inflammatory myalgia[5]	52	19	8
Osteoarthritis[5]	40	13	8
Lyme disease[4‡]	20	NA	15
Hepatitis C infection (no cryoglobulinemia)[23]	50	44	0
HCV-related cryoglobulinemia[23]	29	76	7

NA, not available.
*The percentages shown are based on the specific references cited.
†RF tested by IgM RF ELISA.
‡First-generation anti-CCP assay.

Table 2. Performance of the Second-generation Anti-CCP Assay and RF for Diagnosis of Rheumatoid Arthritis

Reference	Patients, N	Sensitivity (%)				Specificity (%)			
		Anti-CCP	RF	Anti-CCP *and* RF	Anti-CCP *or* RF	Anti-CCP	RF	Anti-CCP *and* RF	Anti-CCP *or* RF
BLOOD DONORS (BEFORE CLINICAL MANIFESTATIONS)									
Berglin et al[12] *	59	37	22	–	–	98	94	–	–
Rantapaa-Dahlqvist et al[10] *	83	34	20	15	–	98	95	99	–
SUSPECTED RHEUMATIC DISEASE									
Sauerland et al[5]	700	74	70	–	–	94	70	–	–
Vallbracht et al[8] *	561	64	66	–	79	97	82	–	81
Greiner et al[14]	333	81	86	–	90	98	82	–	81
De Rycke et al[15]	315	75	78	–	–	97	81	–	–
VARIETY OF RHEUMATIC DISEASES									
Lee et al[6]	249	66	72	57	81	90	80	91	80
RA AT VARIOUS STAGES									
Dubucquoi[16]*	140	64	60	–	75	96	~70	–	–
Suzuki et al[17]	549	88	70	–	–	89	82	–	–
EARLY (<1 YEAR) RA									
Forslind et al[18]	379	55	61	–	–	–	–	–	–
Suzuki et al[17]	91	84	78	–	–	–	–	–	–
LONG-STANDING (≥4 YEARS) RA									
De Rycke et al[15†]	180	66	25	–	–	–	–	–	–

CCP, cyclic citrullinated peptide; RF, rheumatoid factor; RA, rheumatoid arthritis.
RF analysis by latex fixation/immunoturbidimetry unless otherwise indicated; –, data not available.
*IgM RF by ELISA.
†Cut-points were adapted to yield ≥98.5% specificity.

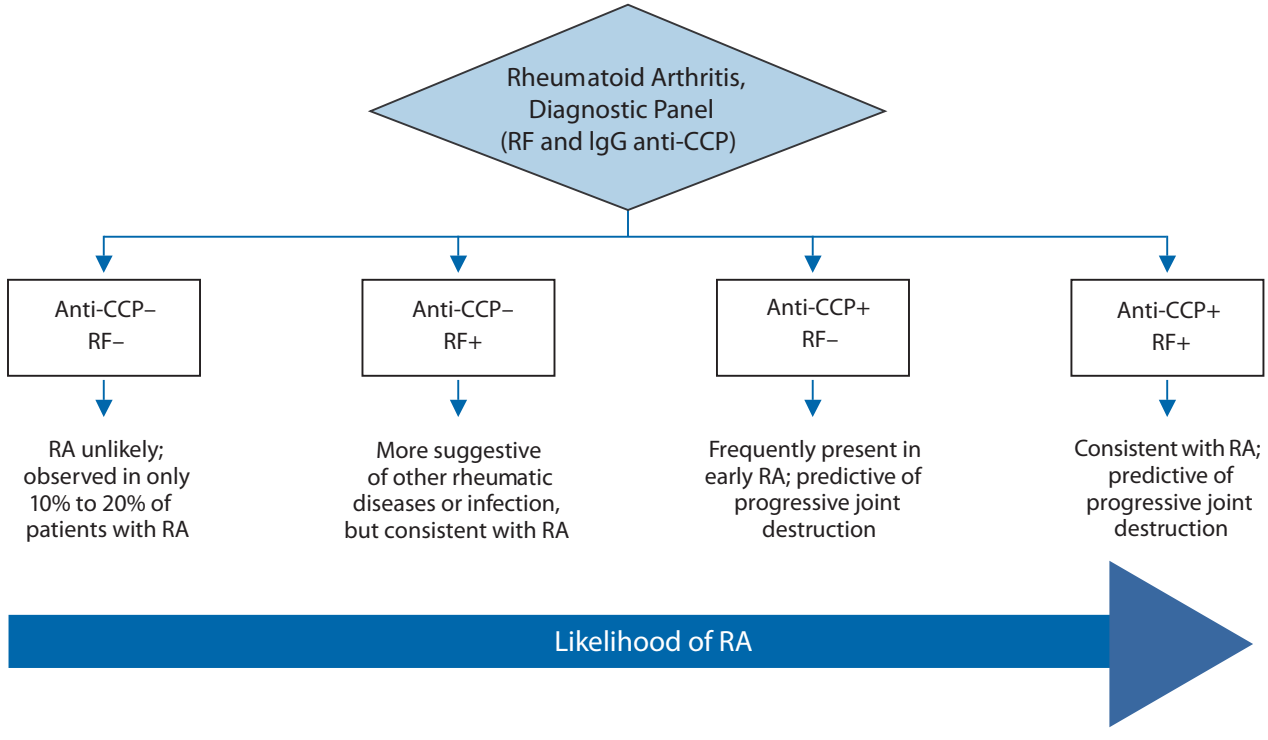

Figure. Flowchart for initial laboratory testing in patients with polyarthritis. Diagnosis should be based on clinical, radiographic, and laboratory findings. See Interpretive Information section for detailed discussion. The arrow indicates the relative likelihood of RA with different combinations of results; however, even patients with negative results on both tests may have RA.

Table 1. Tests Available to Support Diagnosis, Prognosis, and Follow-up of Rheumatoid Arthritis

Assay	Method	Clinical Use
Rheumatoid Factor (RF)	Latex fixation/immunoturbidimetry	Most widely used test to assist in diagnosis and determining prognosis of RA; detects primarily IgM RF
Rheumatoid Factor (IgM)	ELISA	May be used in place of latex fixation/agglutination method, especially if another RF isotype is to be tested
Rheumatoid Factor (IgA)	ELISA	Provides added specificity when used in combination with other RF or anti-CCP assays
Rheumatoid Factor (IgG)	ELISA	Provides added specificity when used in combination with other RF or anti-CCP assays
Anti-Cyclic Citrullinated Peptide (anti-CCP)	ELISA	Assist in diagnosis and determining prognosis of RA—more specific than RF
Rheumatoid Arthritis, Diagnostic Panel RF Anti-CCP	 Latex fixation/immunoturbidimetry ELISA	Provides additional diagnostic and prognostic value relative to either assay alone
Erythrocyte Sedimentation Rate (ESR)	Modified Westergren	Assess disease activity
C-Reactive Protein (CRP)	Chemiluminescent Immunoassay	Assess disease activity

ELISA, enzyme-linked immunosorbent assay.

Until the introduction of the anti-CCP assay (see below), RF was considered the most useful laboratory indicator of RA. The reported sensitivity of RF for RA generally ranges from 60% to 90%, but specificity is relatively low (~70%-80%). Patients with other rheumatic diseases or conditions that present with polyarthritis often have positive RF results. The presence of IgG or IgA RF, or both, in patients with IgM RF and joint disease markedly increases the likelihood that the patient has RA; these combinations are not typically found in patients with other rheumatic diseases that may be accompanied by IgM RF. However, IgA and IgG RF are not highly sensitive and are not widely used in the diagnosis of RA.

Anti-CCP has emerged as a sensitive (~55%-80%) and highly specific (~90%-98%) marker of RA (Table 2).[4,5,6] The data presented in this clinical focus are based on studies using the second-generation anti-CCP immunoassay, which has improved sensitivity and equivalent specificity relative to first-generation assays.[7] In most side-by-side comparisons, anti-CCP is as sensitive as and more specific than RF in various clinical situations (Table 2).

The combination of anti-CCP and RF appears to provide greater sensitivity than either assay alone (Table 2)[8,9] and is therefore useful in the diagnostic work-up of suspected RA (Figure). Notably, in a study of 561 patients with suspected RA, anti-CCP was detected in 35% of RA patients who had negative results for all 3 RF isotypes (IgM, IgA, and IgG by ELISA).[8]

RF and especially anti-CCP can be detected years before the onset of symptoms. In studies of blood donors, the sensitivity of anti-CCP detection for future development of RA ranged from 29% to 37%, with a specificity of ≥98%.[10,11,12] Sensitivity increased as the time to disease onset decreased. Anti-CCP testing may also predict a future diagnosis of RA in patients with a diagnosis of undifferentiated arthritis (UA).[13]

Prognosis

RA has a variable clinical course. Some patients have self-limiting disease whereas others develop progressive joint damage. Predicting which patients will experience progressive disease may help direct aggressive treatment with DMARDs to patients who need it most, and spare others from unnecessary exposure to the potential adverse effects of these drugs.

The presence of RF is generally associated with more severe disease and greater risk of progressive joint erosion. Very high titers may be associated with more severe joint disease, Felty's syndrome, rheumatoid nodules, peripheral neuropathy, and skin ulcers. IgA and IgG RF positivity early in the course of RA is also predictive of more severe disease and likelihood of radiographic progression.[19,20]

Anti-CCP and RF (Figure)

The combination of a positive anti-CCP and IgM RF result is highly specific for RA (~90%-100%) and is associated with an aggressive disease course. However, this profile may be found in some patients with other rheumatic diseases, such as SLE, scleroderma, and psoriatic arthritis. Patients with positive anti-CCP and *negative* RF results are also likely to have RA. RA is less likely in patients with a positive RF and negative anti-CCP result, but cannot be ruled out. Negative results on both assays indicate a very low likelihood of RA, but do not exclude the diagnosis. RF is also associated with extra-articular manifestations such as rheumatoid nodules, whereas anti-CCP is not. In RF-positive patients with chronic HCV or other infections associated with polyarticular arthritis, a positive anti-CCP result suggests a likely diagnosis of RA; however, some HCV patients with cryoglobulinemia may have positive anti-CCP results but do not have RA. [23]

CRP and ESR

Elevated levels of ESR and CRP in patients with RA suggest heightened disease activity. However, elevations may also be due to other inflammatory conditions. Normal ESR and CRP results indicate relatively low disease activity. In patients with discordant ESR and CRP results, CRP levels may be the more reliable marker of RA disease activity. [24] Recent evidence suggests that CRP levels during early RA may be predictive of long-term (10-year) disease progression. [20]

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