Tests for Autoimmune Diseases

Test Codes 249, 16814, 19946

Frequently Asked Questions

Panel components may be ordered separately. Please see the Quest Diagnostics Test Center for ordering information.

1.   Q: What are autoimmune diseases?

A: "Autoimmune disease" refers to a diverse group of disorders that involve almost every one of the body's organs and systems. It encompasses diseases of the nervous, gastrointestinal, and endocrine systems, as well as skin and other connective tissues, eyes, blood, and blood vessels. In all of these autoimmune diseases, the underlying problem is "autoimmunity"—the body's immune system becomes misdirected and attacks the very organs it was designed to protect.

2.   Q: Why are autoimmune diseases challenging to diagnose?

A: Diagnosis is challenging for several reasons:

1. Patients initially present with nonspecific symptoms such as fatigue, joint and muscle pain, fever, and/or weight change.

2. Symptoms often flare and remit.

3. Patients frequently have more than 1 autoimmune disease.

According to a survey by the Autoimmune Diseases Association, it takes up to 4.6 years and nearly 5 doctors for a patient to receive a proper autoimmune disease diagnosis.[1]

3.   Q: How common are autoimmune diseases?

A: At least 30 million Americans suffer from 1 or more of the 80 plus autoimmune diseases.

On average, autoimmune diseases strike three times more women than men. Certain ones have an even higher female:male ratio. Autoimmune diseases are one of the top 10 leading causes of death among women age 65 and under[2] and represent the fourth-largest cause of disability among women in the United States.[3] Women's enhanced immune system increases resistance to infection, but also puts them at greater risk of developing autoimmune disease than men.

Autoimmune disease commonly occurs in multiple members of a family, indicating a genetic predisposition. Family members are affected by various autoimmune disorders rather than one specific disorder.

4.   Q: What is the first test to be considered for a patient suspected of having an autoimmune disease?

A: When evaluating a patient for autoimmune disease, an antinuclear antibody (ANA) test is typically performed first. The immunofluorescence assay (IFA) (test code 249) screens for approximately 150 autoantibodies, which can occur in various autoimmune diseases. The American College of Rheumatology (ACR) recommends IFA as the gold standard method for ANA testing.[4]

A negative ANA IFA result suggests ANA-associated autoimmune diseases are not present. A positive result suggests the presence of autoimmune disease, and reflexes to titer and pattern. A low ANA titer (1:40 to 1:80) is consistent with preclinical disease or lack of disease. Titers >1:80 are consistent with autoimmune disease. In these cases, the staining pattern helps predict the disease type; however, specific antibody testing can be considered useful if clinically indicated.

5. Q: What is the significance of ANA patterns?

A: The significance of various patterns is shown below.

Antibody Pattern	Interpretation
Nuclear Patterns	
Nuclear membrane (nuclear laminae, rim)	Associated with autoimmune liver disease, including primary biliary cirrhosis and autoimmune hepatitis; also associated with SLE, Sjögren syndrome, and seronegative arthritis
Centromere	Associated with the CREST syndrome[a] or Raynaud[a] syndrome
Homogenous	Consistent with presence of antibodies to native DNA, histones, and/or deoxyribonucleoprotein; associated with SLE and drug-induced lupus
Nucleolar	Suggestive of systemic sclerosis (scleroderma), SLE, Sjögren syndrome, polymyositis, overlap syndromes, or Raynaud phenomenon
Proliferating cell nuclear antigen (PCNA)	Highly specific for SLE
Speckled	Suggestive of antibodies to RNP, SS-A, SS-B, Sm, centromere, p95 or p80 coilin; associated with mixed connective tissue disease, SLE, Sjögren syndrome, dermatomyositis, and systemic sclerosis/polymyositis overlap
Nuclear dots (1-6 per cell)	Consistent with Sjögren syndrome, SLE, systemic sclerosis, polymyositis, and asymptomatic individuals
Nuclear dots (6-20 per cell)	Consistent with primary biliary cirrhosis, polymyositis/dermatomyositis, and other systemic autoimmune rheumatic diseases
Cytoplasmic Patterns	
Cytoskeletal	Associated with autoimmune liver disease (anti-smooth muscle); myasthenia gravis, Crohn disease, and long-term hemodialysis; alcoholic liver disease, rheumatoid arthritis, and psoriasis (anti-keratin)
Golgi apparatus	Consistent with SLE, Sjögren syndrome, cerebellar disease, and viral infections
Lysosomal	Unknown clinical significance
Mitochondrial	Suggestive of antimitochondrial antibody presence and primary biliary cirrhosis
Ribosomal	Unknown clinical significance; may be associated with neuropsychiatric lupus

SLE, systemic lupus erythematosus; RNP antibody, ribonucleoprotein antibody; SS-A, SS-B antibodies, Sjögren syndrome antibodies A and B; and Sm antibody, Smith antibody.

[a] CREST is a syndrome defined by presence of calcinosis, Raynaud phenomenon, esophageal dysmotility, sclerodactyly, and telangiectasia.

Note: At Quest Diagnostics each pattern is reported if more than one is observed.

![Quest Diagnostics logo]

6. Q: Can I use one patient specimen to both screen and test for specific antibodies?

A: Yes, Quest Diagnostics offers test code 16814 (ANA Screen, IFA, Reflex Titer/Pattern and Reflex to Multiplex 11 Ab Cascade). This test begins with an ANA screen using IFA technology. A positive result reflexes to titer and pattern and to a 3-tiered, 11-antibody cascade. The first tier includes chromatin, dsDNA, RNP, Sm, and Sm/RNP antibodies. If all 5 antibodies are negative, testing proceeds to the second tier, which includes Jo-1, Scl-70, SS-A, and SS-B antibodies. If all 4 of these antibodies are negative, testing proceeds to the final tier, which includes centromere B and ribosomal P antibodies.

Figure 1 details the cascade and interpretation of specific antibody results. Note that if the ANA IFA result is positive but all 11 specific antibody results are negative, an autoimmune disease may still be present. The disease may be associated with an antibody not tested for in the cascade. Diseases to be considered include rheumatoid arthritis, autoimmune hepatitis, primary biliary cirrhosis, autoimmune thyroiditis, Addison disease, pernicious anemia, autoimmune neuropathies, vasculitis, celiac disease, bullous disease, and others.

7. Q: Can I order specific antibody testing without an ANA (IFA) screen?

A: The ANA Multiplex with Reflex to 11 Antibody Cascade (test code 19946) can be used. This test does not include an ANA screen based on IFA technology. This less sensitive, but more specific test uses multiplex bead immunoassay technology. The test includes a 3-tiered, 11-antibody cascade that begins with chromatin, dsDNA, RNP, Sm, and Sm/RNP antibodies. If all 5 antibodies are negative, testing proceeds to the second tier, which includes Jo-1, Scl-70, SS-A, and SS-B antibodies. If all 4 of these antibodies are negative, testing proceeds to the final tier, which includes centromere B and ribosomal P antibodies.

A negative result on all 11 antibodies does not rule out autoimmune disease. A disease associated with an antibody not tested in the cascade may be present, especially if the patient has previously tested positive on an ANA IFA screen. Diseases to be considered include rheumatoid arthritis, autoimmune hepatitis, primary biliary cirrhosis, autoimmune thyroiditis, Addison disease, pernicious anemia, autoimmune neuropathies, vasculitis, celiac disease, bullous disease, and others.

8. Q: How predictive are the specific antibodies? What is their sensitivity and specificity in various autoimmune diseases?
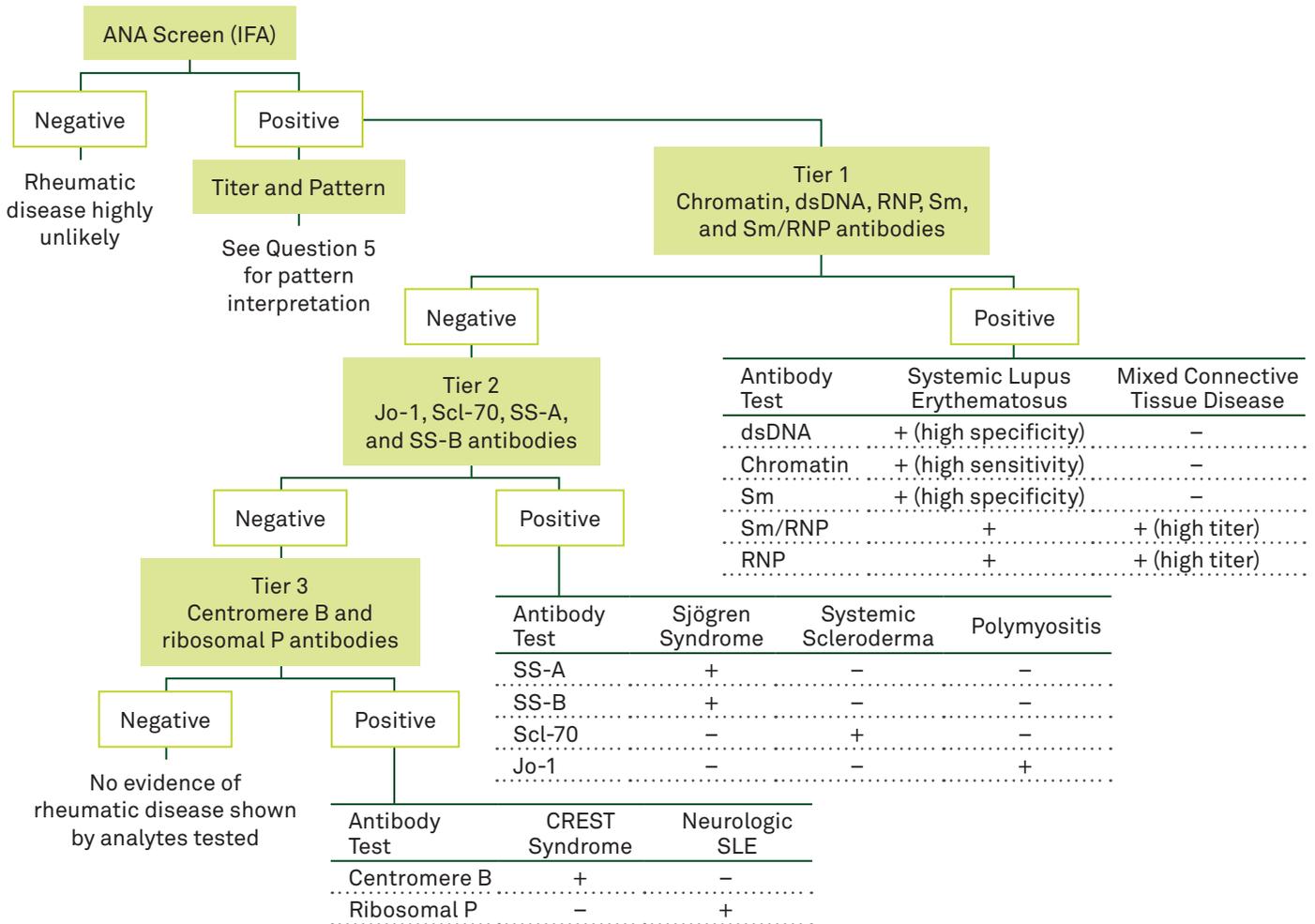
A: Presence of a specific antibody is highly suggestive for the associated autoimmune disease. However, these antibodies are not specific for a particular disease; thus, results need to be interpreted in context of the clinical information and the following antibody prevalence.

Prevalence of Tier 1 Antibodies[5-8]
- *Double-stranded DNA (dsDNA) antibodies* are present in 57% to 62% of systemic lupus erythematosus (SLE) cases, 10% to 43% of polymyositis, 11% to 20% of Sjögren syndrome, 8% of systemic sclerosis (scleroderma), and 0% to 8% of mixed connective tissue disease (MCTD).
- *Chromatin antibody* is present in >80% of MCTD cases, 37% to 73% of SLE, 14% of systemic sclerosis, 12% of Sjögren syndrome, and 8% of polymyositis.
- *Ribonucleoprotein (RNP) antibodies* target RNP A and/or RNP 68kD proteins; antibodies to one or both are present in >80% of MCTD cases, 22% to 48% of SLE, 14% of systemic sclerosis, 12% of Sjögren syndrome, and 8% of polymyositis.

Figure 1. Use of ANA (IFA) and Specific Antibody Testing Cascade (Test Code 16814) for the Diagnosis of Rheumatic Disease

ANA Screen (IFA)

- Negative → Rheumatic disease highly unlikely
- Positive
  - Titer and Pattern → See Question 5 for pattern interpretation
  - Tier 1 Chromatin, dsDNA, RNP, Sm, and Sm/RNP antibodies
    - Negative
      - Tier 2 Jo-1, Scl-70, SS-A, and SS-B antibodies
        - Negative
          - Tier 3 Centromere B and ribosomal P antibodies
            - Negative → No evidence of rheumatic disease shown by analytes tested
            - Positive
        - Positive
    - Positive

Antibody Test	Systemic Lupus Erythematosus	Mixed Connective Tissue Disease
dsDNA	+ (high specificity)	−
Chromatin	+ (high sensitivity)	−
Sm	+ (high specificity)	−
Sm/RNP	+	+ (high titer)
RNP	+	+ (high titer)

Antibody Test	Sjögren Syndrome	Systemic Scleroderma	Polymyositis
SS-A	+	−	−
SS-B	+	−	−
Scl-70	−	+	−
Jo-1	−	−	+

Antibody Test	CREST Syndrome	Neurologic SLE
Centromere B	+	−
Ribosomal P	−	+

The acronym CREST refers to a syndrome defined by presence of calcinosis, Raynaud phenomenon, esophageal dysmotility, sclerodactyly, and telangiectasia. dsDNA indicates double-stranded DNA; Sm/RNP antibody, Smith/ribonucleoprotein antibody; SS-A and -B antibodies, Sjögren syndrome A and B antibodies; Scl-70 antibody, scleroderma (topoisomerase I) antibody; Jo-1 antibody, histidyl-tRNA synthetase antibody; and SLE, systemic lupus erythematosus.

This figure was developed by Quest Diagnostics based on references 1-5. It is provided for informational purposes only and is not intended as medical advice. A physician's test selection and interpretation, diagnosis, and patient management decisions should be based on his/her education, clinical expertise, and assessment of the patient.

References

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8. Q: How predictive are the specific antibodies? What is their sensitivity and specificity in various autoimmune diseases? (continued)

- *Sm/RNP antibodies* are directed to epitopes formed in a complex of Sm and RNP; antibodies to the Sm/RNP complex are present in 54% to 94% of MCTD cases, 30% of SLE, 4% of systemic sclerosis, and 9% of Sjögren syndrome and polymyositis.

- *Sm antibody* is present in 20% to 30% of SLE cases, 8% of MCTD, 10% of polymyositis, 0% of systemic sclerosis, and 4% of Sjögren syndrome.

Double stranded DNA, chromatin, ribonucleoprotein, Sm/RNP complex and Sm antibodies are also present in <2% of normal blood donors.

Prevalence of Tier 2 Antibodies[7,8]

- *SS-A and SS-B antibodies* are present in >80% of Sjögren syndrome cases and are considered a diagnostic indicator for this autoimmune disease. However, these antibodies are also present in other autoimmune disorders. SS-A antibodies are seen in 33% to 52% of SLE cases, 42% of polymyositis, 23% of systemic sclerosis (scleroderma), and 13% of MCTD. SS-B antibody is present in 13% to 27% of SLE cases, 5% of systemic sclerosis, <2% of polymyositis, and <2% of MCTD.

- *Scl-70 antibody* is present in 16% of systemic sclerosis cases, 7% of MCTD (especially those with features of systemic sclerosis), 2% to 3% of SLE, <2% of Sjögren syndrome, and <2% of polymyositis.

- *Jo-1 antibody* is present in 17% of polymyositis cases, 7% of MCTD (especially in those with features of muscle inflammation), <2% of SLE, Sjögren syndrome, and systemic sclerosis.

Tier 2 antibodies are also present in <2% of normal blood donors.

Prevalence of Tier 3 Antibodies[7-10]

- *Centromere B antibody* is present in 27% of systemic sclerosis (scleroderma) cases, 66% of CREST syndrome (calcinosis, Raynaud phenomenon, esophageal dysmotility, sclerodactyly, and telangiectasia), 3% to 12% of SLE, 7% of MCTD (typically with features of polymyositis), and <2% of Sjögren syndrome, polymyositis, and normal blood donors.

- *Ribosomal P antibody* is present in 9% to 30% of SLE cases (often with neurological manifestations), 7% of MCTD, and <2% of Sjögren syndrome, systemic sclerosis, polymyositis, and normal blood donors.

Note
The prevalences listed above may vary with the population studied and methods used.[5] Values given are for general guidance.

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