MAYO CLINIC LABORATORIES

# Test Definition: FACT

Filamentous-Actin (F-actin) Antibody, IgG, Serum

## Overview

### Useful For

Evaluation of patients with hepatitis of unknown origin associated with hypergammaglobulinemia and/or abnormal liver enzymes

This test may also be useful for confirming positivity for smooth muscle antibodies.

### Testing Algorithm

For more information see First-Line Screening for Autoimmune Liver Disease Algorithm.

### Special Instructions

- First-Line Screening for Autoimmune Liver Disease Algorithm

### Method Name

Enzyme-Linked Immunosorbent Assay (ELISA)

### NY State Available

Yes

## Specimen

### Specimen Type

Serum

### Specimen Required

**Supplies:** Sarstedt Aliquot Tube, 5 mL (T914)
**Collection Container/Tube:**
**Preferred:** Serum gel
**Acceptable:** Red top
**Submission Container/Tube:** Plastic vial
**Specimen Volume:** 0.5 mL
**Collection Instructions:** Centrifuge and aliquot serum into a plastic vial.

### Specimen Minimum Volume

0.4 mL

### Reject Due To

| | |
|---|---|
| Gross | OK |

| | |
|---|---|
| hemolysis | |
| Gross lipemia | OK |
| Gross icterus | OK |
| Heat treated specimens | Reject |

### Specimen Stability Information

| Specimen Type | Temperature | Time | Special Container |
|---|---|---|---|
| Serum | Refrigerated (preferred) | 21 days | |
| | Frozen | 21 days | |

## Clinical & Interpretive

### Clinical Information

Autoimmune hepatitis (AIH) is a chronic disease resulting from immune-mediated liver injury with varied clinical manifestations.(1,2) The precise factors leading to disease initiation and perpetuation are unknown, but likely reflect a combination of genetic predisposition relating to defects in immunological control of autoreactivity, as well as environmental triggers, which precipitate a persistent breakdown in self-tolerance.(2) Initially, patients with AIH may be clinically asymptomatic and are usually identified only through an incidental finding of abnormal liver function tests.(1-4) At a more advanced stage, patients may manifest with symptoms such as jaundice, pruritus, or ascites, which are secondary to the more extensive liver damage. As implied by the name, AIH has many characteristics of an autoimmune disease, including female predominance, hypergammaglobulinemia, association with specific *HLA* alleles, responsiveness to immunosuppression, and the presence of autoantibodies.(1-3)

The clinical features of AIH are nonspecific and can be seen in variety of liver diseases (drug/alcohol-associated hepatitis, viral hepatitis, primary sclerosing cholangitis, etc), the diagnosis can be challenging. A set of diagnostic criteria for AIH has been published and includes the presence of various autoantibodies, elevated total IgG, evidence of hepatitis on liver histology, and absence of viral markers.(1,3,4) Based on the specific autoantibodies present, AIH can be placed into one of three categories.(4) The most prevalent is AIH type 1, linked to the presence of smooth muscle autoantibodies (SMA), antinuclear antibodies (ANA) and perinuclear anti-neutrophil cytoplasmic antibodies. SMA are generally identified by indirect immunofluorescence using a smooth muscle substrate. The antigen specificity of SMA in the context of AIH has been identified as filamentous-actin (F-actin).(3) The combination of autoantibody serology, specifically SMA and anti-F-actin antibodies with liver histology and thorough clinical evaluation are useful in the evaluation of patients with suspected autoimmune hepatitis. SMAs are detected in up to 85% of patients with AIH, either alone or in conjunction with ANA.(1,4,5) The SMA titer can also contribute to International Autoimmune Hepatitis Group diagnostic score in patients with a probable or definite diagnosis of AIH.(1,4,5) These antibodies have also been reported in 33% to 65% of cases of primary biliary cholangitis/AIH overlap syndrome,(6) the concomitant presence of SMA and antimitochondrial antibodies being highly suggestive in this setting.

For more information see First-Line Screening for Autoimmune Liver Disease Algorithm.

## Reference Values

Negative: <20.0 U
Weak Positive: 20.0-30.0 U
Positive: >30.0 U

## Interpretation

Positivity for anti-filamentous-actin (F-actin) antibodies may help support a diagnosis of autoimmune hepatitis (AIH) following exclusion of other causes of hepatitis.

A negative result for anti-F-actin antibodies does not exclude a diagnosis of AIH.

In a study conducted at Mayo Clinic, the F-actin enzyme-linked immunosorbent assay (ELISA) had a clinical sensitivity of 92.9% when using the manufacturer's recommended cutoff of 20.0 U. In addition, the F-actin ELISA had a clinical specificity of 76.7% when using the aforementioned cutoffs. See Supportive Data.

## Cautions

Serologic tests for autoantibodies, including anti-filamentous-actin (F-actin), should not be relied upon exclusively to determine the etiology or prognosis of patients with liver disease.

A negative result for anti-F-actin antibodies does not exclude a diagnosis of autoimmune hepatitis.

## Supportive Data

In a study performed at Mayo Clinic, 173 serum samples submitted for clinical testing for anti-smooth muscle antibodies (anti-SMA), as performed by indirect immunofluorescence, were collected. These samples were subsequently tested using the anti-filamentous-actin (F-actin) antibody enzyme-linked immunosorbent assay (ELISA). By using the manufacturer's cut-offs for the 2 tests (negative at <20.0 units for the F-actin ELISA and <1:20 titer for the anti-SMA indirect immunofluorescence), the 2 tests had an overall concordance of 79.8%. In addition to the analytical concordance, patient histories were abstracted for diagnoses related to liver dysfunction. Of the 14 patients with autoimmune hepatitis, 13 were positive (> or =20.0 units) for F-actin antibodies by ELISA, which corresponded to a sensitivity of 92.9%. Of the remaining 159 patients who had a diagnosis of something other than autoimmune hepatitis, 122 were negative (<20.0 units), which corresponded to a specificity of 76.7%. In comparison, at a clinical specificity of 76.1%, which is similar to the ELISA, the anti-SMA indirect immunofluorescence method had a significantly lower clinical sensitivity of 78.6%. Positivity for either anti-F-actin antibodies or anti-SMA improved the diagnostic sensitivity to 92.9%, although the specificity decreased to 66.0%. This data indicates that the ELISA for F-actin antibodies may have improved diagnostic utility in comparison to the anti-SMA by indirect immunofluorescence, although a combination of these tests may be useful for some patients.

## Clinical Reference

1. European Association for the Study of the Liver. EASL clinical practice guidelines: autoimmune hepatitis. J Hepatol. 2015;63(4):971-1004
2. Mieli-Vergani G, Vergani D, Czaja AJ, et al. Autoimmune hepatitis. Nat Rev Dis Primers. 2018;4:18017
3. Sebode M, Weiler-Normann C, Liwinski T, Schramm C. Autoantibodies in autoimmune liver disease-clinical and diagnostic relevance. Front Immunol. 2018;9:609
4. Terziroli Beretta-Piccoli B, Mieli-Vergani G, Vergani D. Autoimmune hepatitis: serum autoantibodies in clinical practice. Clin Rev Allergy Immunol. 2022;63(2):124-137

5. Bogdanos DP, Invernizzi P, Mackay IR, Vergani D. Autoimmune liver serology: current diagnostic and clinical challenges. World J Gastroenterol. 2008;14(21):3374-3387
6. Muratori P, Granito A, Pappas G, et al. The serological profile of the autoimmune hepatitis/primary biliary cirrhosis overlap syndrome. Am J Gastroenterol. 2009;104(6):1420-1425.

## Performance

### Method Description

The method used to detect antibodies directed against filamentous-actin is enzyme-linked immunosorbent assay. Prediluted controls and diluted patient sera are added to separate wells, allowing any actin antibodies present to bind to the antigen. Unbound sample is washed away, and an enzyme labeled anti-human IgG is added to each well. A second incubation allows the enzyme labeled anti-human IgG to bind to any patient antibodies, which have become attached to the microwells, and any unbound conjugate is removed by another wash step. The bound conjugate is visualized with 3,3',5,5' tetramethylbenzidine (TMB) substrate, which gives a blue reaction product, the intensity of which is proportional to a concentration of autoantibody in the sample. Sulfuric acid is added to each well to stop the reaction. This produces a yellow endpoint color, which is read at 450 nm. Testing is performed on the DS2 instrument by Dynex.(Package insert: QUANTA Lite Actin IgG ELISA 708785. INOVA Diagnostics; Rev. 6, 11/2018)

### PDF Report

No

### Day(s) Performed

Monday

### Report Available

2 to 8 days

### Specimen Retention Time

14 days

### Performing Laboratory Location

Mayo Clinic Laboratories - Rochester Superior Drive

## Fees & Codes

### Fees

- Authorized users can sign in to Test Prices for detailed fee information.
- Clients without access to Test Prices can contact Customer Service 24 hours a day, seven days a week.
- Prospective clients should contact their account representative. For assistance, contact Customer Service.

### Test Classification

# Test Definition: FACT

Filamentous-Actin (F-actin) Antibody, IgG, Serum

This test has been cleared, approved, or is exempt by the US Food and Drug Administration and is used per manufacturer's instructions. Performance characteristics were verified by Mayo Clinic in a manner consistent with CLIA requirements.

## CPT Code Information

83516

## LOINC® Information

| Test ID | Test Order Name | Order LOINC® Value |
|---|---|---|
| FACT | F-Actin Ab, IgG, S | 44706-0 |

| Result ID | Test Result Name | Result LOINC® Value |
|---|---|---|
| FACT | F-Actin Ab, IgG, S | 44706-0 |